AMERICAN ZOOLOGIST

Volume 1, Number 3 August 1961

TABLE OF CONTENTS

Page

American Society of Zoologists: Officers 310

Synopsis of Program 311

Program 314

Abstracts 337

Index to Abstracts 401

AMERICAN SOCIETY OF ZOOLOGISTS

OFFICERS

President-Elect:	Curt Stern University of California Berkeley, Calif.	President:	C. Ladd Prosser University of Illinois Urbana, Ill.
Past-President:	Emil Witschi State University of Iowa Iowa City, Iowa	Secretary:	Charles B. Metz Florida State University Tallahassee, Fla.
Treasurer:	Jerry J. Kollros State University of Iowa Iowa City, Iowa	Program Officer:	Ray L. Watterson Northwestern University Evanston, Ill.

DIVISIONAL OFFICERS

ANIMAL BEHAVIOR

Chairman:	William C. Young University of Kansas Lawrence, Kan.
Vice-Chairman:	William C. Dilger Cornell University Ithaca, N. Y.
Secretary:	Martin W. Schein Pennsylvania State Univ. University Park, Pa.

VERTEBRATE MORPHOLOGY

Chairman:	D. Dwight Davis Chicago Nat. Hist. Mus. Chicago, Ill.
Vice-Chairman:	Harvey Fisher Southern Illinois Univ. Carbondale, Ill.
Secr.-Treas.:	Howard Evans Cornell University Ithaca, N. Y.

COMPARATIVE PHYSIOLOGY

Chairman:	Theodore H. Bullock University of California Los Angeles, Calif.
Chairman-Elect:	Arthur D. Hasler University of Wisconsin Madison, Wisc.
Program Officer:	Kenneth D. Roeder Tufts College Medford, Mass.
Secr.-Treas.:	Frank J. Vernberg Duke University Durham, N. C.

DEVELOPMENTAL BIOLOGY

Chairman:	James D. Ebert Carnegie Institution Baltimore, Md.
Vice-Chairman:	John A. Moore Columbia University New York, N. Y.
Program Officer:	John W. Saunders Marquette University Milwaukee, Wisc.
Secr.-Treas.:	Beatrice Mintz Institute for Cancer Research Philadelphia, Pa.

COMPARATIVE ENDOCRINOLOGY

Chairman:	Aubrey Gorbman Barnard College New York, N. Y.
Chairman-Elect:	Berta V. Scharrer Albert Einstein College of Med. New York, N. Y.
Secr.-Treas.	Howard A. Bern University of California Berkeley, Calif.
Program Officer:	Paul A. Wright University of New Hampshire Durham, N. H.

INVERTEBRATE ZOOLOGY

Chairman:	Clarence J. Goodnight Purdue University Lafayette, Ind.
Vice-Chairman:	Bertil G. Anderson Pennsylvania State University University Park, Pa.
Program Officer:	Melbourne R. Carriker Fish and Wildlife Service Biological Laboratory Oxford, Maryland
Secr.-Treas.:	Charles F. Lytle Tulane University New Orleans, La.

Other officers, committees, and representatives are listed in *American Zoologist*, Vol. 1, No. 2 (May, 1961).

SYNOPSIS OF PROGRAM

Page

SATURDAY AFTERNOON, AUGUST 26

Meeting: Policy Committee, American Society of Zoologists 315

SATURDAY EVENING, AUGUST 26

Meeting: Executive Committee, American Society of Zoologists 315

SUNDAY MORNING, AUGUST 27

Meeting: Intersociety Officers 315

SUNDAY AFTERNOON, AUGUST 27

Meeting: Executive Committee, American Society of Zoologists 315

MONDAY MORNING, AUGUST 28

Refresher Course: Comparative Neurophysiology, I 316

Section A: Animal Behavior and Sociobiology, I 316

Section B: Comparative Physiology, I 317

Section C: Protozoology 317

MONDAY AFTERNOON, AUGUST 28

Refresher Course: Comparative Neurophysiology, II 318

Section D: Animal Behavior and Sociobiology, II 319

Section E: Comparative Physiology, II 319

Section F: Invertebrate Zoology 320

Business Meeting: Division of Animal Behavior and Sociobiology 319

SYNOPSIS OF PROGRAM

Page

MONDAY EVENING, AUGUST 28

Refresher Course: Comparative Neurophysiology, III 320
Business Meeting: Division of Invertebrate Zoology 320
General Meeting for all Societies 320

TUESDAY MORNING, AUGUST 29

Symposium: Comparative Physiology of Excretory Organs, I 321
Section G: Animal Behavior and Sociobiology, III 321
Section H: Histology and Cytology 322
Section I: Experimental Biology 322
Section J: Developmental Biology, I 323
Luncheon: Division of Animal Behavior and Sociobiology 324

TUESDAY AFTERNOON, AUGUST 29

Symposium: Comparative Physiology of Excretory Organs, II 324
Section K: Animal Behavior and Sociobiology, IV 324
Section L: Comparative Physiology, III 325
Section M: Comparative Endocrinology, I 325
Section N: Developmental Biology, II 325
Tea: Division of Invertebrate Zoology 326
Interim Business Meeting: American Society of Zoologists 326

TUESDAY EVENING, AUGUST 29

Section O: Developmental Biology, III 326
Business Meeting: Division of Comparative Physiology 326

WEDNESDAY MORNING, AUGUST 30

Symposium: Origin, Structure and Functioning of Hemocytes in Invertebrates, I 327
Section P: Developmental Biology, IV 327
Section Q: Behavioral Films, I. Animal Behavior and Sociobiology, V 328
Section R: Comparative Physiology, IV 329
Section S: Comparative Endocrinology, II 329
Luncheon: Division of Developmental Biology 330

Page

WEDNESDAY AFTERNOON, AUGUST 30

Symposium: Origin, Structure and Functioning of Hemocytes in Invertebrates, II 330
Section T: Developmental Biology, V 330
Section U: Behavioral Films, II. Animal Behavior and Sociobiology, VI 331
Section V: Demonstration Film 331
Section W: Comparative Endocrinology, III 331
Business Meeting: Division of Comparative Endocrinology 332

WEDNESDAY EVENING, AUGUST 30

Zoologists' Dinner and Presidential Address 332

PROGRAM

Summer Meeting of the American Society of Zoologists

August 28 through 30, 1961

Purdue University, Lafayette, Indiana

A brief synopsis of the program is given on pages 311-313 of this issue. 192 papers will be read in 29 sessions including the refresher course and the two 2-session symposia. 82 papers will be presented by title. Dr. M. X. Zarrow is our local representative.

Refresher Course. The topic of the sixth of these special courses is *Comparative Neurophysiology.* The course has been organized by Dr. Donald Kennedy and has received financial support from the National Science Foundation. The National Association of Biology Teachers co-sponsors this program.

Animal Behavior and Sociobiology. This division has arranged 4 sessions of contributed papers and 2 sessions of behavioral films all co-sponsored by the Section of Animal Behavior and Sociobiology of the Ecological Society of America. A luncheon and business meeting will be held.

Comparative Endocrinology. Three sessions of contributed papers have been arranged. A business meeting will be held.

Comparative Physiology. A 2-session symposium on *Comparative Physiology of Excretory Organs* has been organized by Dr. Leonard B. Kirschner. Four sessions of contributed papers will be presented. A business meeting will be held.

Developmental Biology. Five sessions of contributed papers will be held. Two are co-sponsored by the Society for the Study of Development and Growth and the Developmental Biology Section of the Botanical Society of America. The other three sessions are co-sponsored only by the former society. A luncheon will be held but no business meeting.

Invertebrate Zoology. A 2-session symposium on *Origin, Structure and Functioning of Hemocytes in Invertebrates* has been organized by Dr. Melbourne R. Carriker. In addition there will be one session of contributed papers, a tea and a business meeting.

Miscellaneous Sessions. Three additional sessions of contributed papers will be held, one devoted to *Protozoology,* one to *Histology and Cytology* and one to *Experimental Biology.* A special 40-minute demonstration film will be presented by Dr. Hans Elias on three dimensional interpretation of flat sections.

Interim Business Meeting of the American Society of Zoologists. This meeting is called for Tuesday afternoon at 5:15 P.M.

Zoologists' Dinner and Presidential Address. Wednesday evening. Dr. C. Ladd Prosser, University of Illinois, will speak on "The Challenge and Responsibility of Zoology."

For the past several meetings chairmen and speakers have cooperated to keep the programs exactly on the published time schedule. This has been an important feature in the success of the meetings and this policy should continue. If a speaker fails to appear, the free time may be used for further discussion or for a brief recess. Any speaker or presiding officer who finds at the last minute that he cannot attend the meeting is urged to notify either the Program Officer of the Division concerned or Ray L. Watterson, Program Officer of the Society.

SATURDAY AFTERNOON, AUGUST 26

Meeting: Policy Committee of the American Society of Zoologists 3:00 P.M.
Lilly Life Sciences Building, Room 1-117

SATURDAY EVENING, AUGUST 26

Meeting: Executive Committee of the American Society of Zoologists 7:30 P.M.
Lilly Life Sciences Building, Room 1-117

SUNDAY MORNING, AUGUST 27

Meeting: Intersociety Officers 10:00 A.M.
Lilly Life Sciences Building, Room 1-117

SUNDAY AFTERNOON, AUGUST 27

Meeting: Executive Committee of the American Society of Zoologists 2:00 P.M.
Lilly Life Sciences Building, Room 1-117

LIST OF TITLES

Papers are arranged in the order of presentation. Those to be presented by title only are listed last. The abstracts, which follow the list of titles, are arranged alphabetically by the first author.

MONDAY MORNING SESSION, AUGUST 28

Refresher Course: *Comparative Neurophysiology*, First Lecture Session 9:00 A.M.

Sponsored by the American Society of Zoologists and the National Association of Biology Teachers. Supported by a grant from the National Science Foundation. Organized by Donald Kennedy. Two sessions of lectures, and one for discussion; demonstrations have also been arranged, and will take place in the Life Sciences Building during the lunch hour and between 5:00 and 8:00 P.M. on August 28 in rooms to be announced. No abstracts published for Refresher Course.

Lilly Life Sciences Building, Room 1-105

C. LADD PROSSER, *presiding*

9:00 1. PROSSER, C. LADD, University of Illinois (President of the American Society of Zoologists). Introduction.
9:15 2. HOYLE, GRAHAM, University of Glasgow. Comparative physiology of conduction in nerve and muscle.
10:00 3. KENNEDY, DONALD, Stanford University. The initiation of impulses in receptors.
10:40 4. FLOREY, ERNST, University of Washington. Recent studies on synaptic transmitters.
11:20 5. VAN DER KLOOT, WILLIAM G., New York University. Muscle and its neural activation.

Section A. *Animal Behavior and Sociobiology, I.* 9:00 A.M.
(In collaboration with the Section of Animal Behavior and Sociobiology of the Ecological Society of America)

Smith Hall, Room 108

N. E. COLLIAS, *presiding*

9:00 6. TAVOLGA, W. N., American Museum of Natural History. Mechanisms of sound production in the marine catfishes, *Galeichthys* and *Bagre*. (15 min.)
9:20 7. SCOTT, J. P., Roscoe B. Jackson Memorial Laboratory. Spectrographic analysis of dog sounds. (15 min.)
9:40 8. PAYNE, R. S., Cornell Universtiy. The acoustical localization of prey by the Barn Owl (*Tyto alba*). (15 min.)
10:00 9. MOORE, R. E., University of Texas. Olfactory discrimination by individuals of *Peromyscus maniculatus* and *Peromyscus polionotus*. (Introduced by W. F. Blair) (15 min.)

10:20 10. DICE, L. R. and J. P. CLARK, University of Michigan, Michigan State University and Cranbrook Institute of Science. Variation in behavior and temperament among three races of peromyscus. (15 min.)

10:40 11. BARBEHENN, K. R., National Institute of Mental Health. Interspecific spatial relationships among small mammals. (15 min.)

11:00 12. BALPH, D. F., Utah State University. The behavioral response of free Uinta ground squirrels (*Citellus armatus*) to a trap. (15 min.)

11:20 13. TERMAN, C. R., Penrose Research Laboratory, Zoological Society, Philadelphia. Behavioral factors associated with the cessation of growth of laboratory populations of Prairie Deermice. (15 min.)

11:40 14. BRONSON, F. H. and D. E. DAVIS, Pennsylvania State University. The effect of density and aggression on interaction rates in woodchuck populations. (15 min.)

Section B. *Comparative Physiology, I.* 9:20 A.M.

Lilly Life Sciences Building, Room G-126

A. W. MARTIN, *presiding*

9:20 15. FOX, SISTER ALICE MARIE, Clarke College. Active transport of sugar by turtle small intestine. (Introduced by X. J. Mussachia) (15 min.)

9:40 16. PRIVITERA, C. A. and H. J. MERSMANN, Saint Louis University. Comparative oxidative phosphorylation by mitochondria from tissues of the turtle, *Chrysemys picta.* (15 min.)

10:00 17. GOLDBERG, ERWIN, VIRGINIA W. PENCE and CHARLES NORMAN, West Virginia University. The distribution of free amino acids in cock and bull seminal plasma and spermatozoa. (15 min.)

10:20 18. HOFFERT, JACK R. and P. O. FROMM, Michigan State University. *In vitro* uptake of hexavalent chromium by RBC, liver and kidney tissue of the turtle. (15 min.)

10:40 19. ZIEGLER, FRED, E. HARDIN STRICKLAND and ADAM ANTHONY, Pennsylvania State University. Oxygen electrode measurement of tissue slice and homogenate respiration. (15 min.)

11:00 20. Paper cancelled. Abstract published anyway.

11:20 21. WILSON, FRANCIS H. and PHILIP N. NIOSI, Lebanon Valley College. Some observations of gastric digestion in the horned owl. (10 min.)

Section C. *Protozoology* 9:00 A.M.

Lilly Life Sciences Building, Room G-431

SALLY LYMAN ALLEN, *presiding*

9:00 22. WIDMAYER, DOROTHEA, Indiana University. The detection of two new mutations of hump kappa in killer stock 51, syngen 4, *Paramecium aurelia.* (Introduced by T. W. Torrey) (15 min.)

9:20 23. SCHNELLER, MYRTLE V., Indiana University. Genic interrelationships between two particle-bearing stocks of syngen 4, *Paramecium aurelia.* (15 min.)

9:40 24. MUELLER, JO ANNE, Indiana University. Further studies on the nature of the "co-factor" required for kappa infection in *Paramecium aurelia,* syngen 4. (15 min.)

10:00 25. SMITH, JOAN EILEEN, Indiana University. Purification of kappa particles of *Paramecium aurelia,* stock 51. (Introduced by Sears Crowell) (15 min.)

10:20 26. BARNETT, AUDREY, Indiana University. The inheritance of mating type and cycling in *Paramecium multimicronucleatum,* syngen 2. (Introduced by F. Payne) (15 min.)

10:40 27. MIYAKE, AKIO, Indiana University and Osaka City University, Osaka, Japan. Artificial induction of conjugation by chemical agents in *Paramecium* of the "*aurelia* group" and some of its applications to genetic work. (Introduced by R. V. Dippell) (15 min.)

11:00 28. ELLIOTT, ALFRED M., JOHN R. KENNEDY, JR. and IL JIN BAK, University of Michigan. Macronuclear events during mitosis in *Tetrahymena pyriformis.* (15 min.)

11:20 29. BARRETT, JAMES M. and SISTER DOMINIC MARIE OLINGER, Marquette University. An immunochemical test of the reliability of nuclear morphology as a taxonomic criterion for actinophryid Protozoa. (15 min.)

11:40 30. KERR, NORMAN S., University of Minnesota. The inhibition of flagella formation by streptomycin. (15 min.)

12:00 31. GRIFFIN, JOE L. and PHILLIP R. SWEENY, Brown University. Physical and chemical studies on the external membranes of *Amoeba, Chaos* and *Pelomyxa.* (15 min.)

MONDAY AFTERNOON SESSION, AUGUST 28

Refresher Course: *Comparative Neurophysiology,* Second Lecture Session

2:00 P.M

Lilly Life Sciences Building, Room 1-105

DONALD KENNEDY, *presiding*

2:00 32. WIERSMA, C. A. G., California Institute of Technology. The organization of arthropod central nervous systems.

2:40 33. MAYNARD, DONALD M., University of Michigan. Invertebrate neuropile.

3:20 34. BULLOCK, T. H., University of California, Los Angeles. Rythmicity and integration in neural systems.

4:00 35. ROEDER, KENNETH D., Tufts University. The neural mechanisms of behavior.

Section D. *Animal Behavior and Sociobiology, II.* 2:00 P.M.
(In collaboration with the Section of Animal Behavior and Sociobiology of the Ecological Society of America)

Smith Hall, Room 108

W. N. TAVOLGA, *presiding*

2:00 36. BOVEE, E. C. and D. E. WILSON, University of Florida. An ameboid stage in the feeding behavior of the helioflagellate protozoan, *Dimorpha floridanis.* (15 min.)

2:20 37. NASH, T. L., J. M. WARREN and M. W. SCHEIN, Pennsylvania State University. Effects of time and interpolated problems upon retention in chickens. (15 min.)

2:40 38. KLOPFER, F. D., Washington State University. Early experience and discrimination learning in swine. (15 min.)

3:00 39. MOORE, A. U. and HELEN BLAUVELT, Cornell University. Behavior during a conditioned reflex training period used as a measure of early trauma in lamb and kid. (15 min.)

3:20 40. BRADDOCK, J. C., Z. I. BRADDOCK and H. RICHTER, Michigan State University. Form discrimination in the Siamese Fighting Fish, *Betta splendens.* (15 min.)

Business Meeting

Section of Animal Behavior and Sociobiology of the American Society of Zoologists and the Ecological Society of America 4:00 P.M.

Smith Hall, Room 108

Section E. *Comparative Physiology, II.* 2:00 P.M.

Lilly Life Sciences Building, Room G-126

BRADLEY SCHEER, *presiding*

2:00 41. MATSUMOTO, KUNIO, Okayama University and University of Oregon. Peripheral secretion of neurosecretory cells in ventral ganglia of decapod crustaceans. (Introduced by B. T. Scheer) (15 min.)

2:20 42. STRAUS, LORNA P. and W. L. DOYLE, University of Chicago. Fine structure of the so-called chloride cells of the gills of the guppy. (15 min.)

2:40 43. FLEMING, WARREN R., KENNETH SCHEFFEL and J. R. LINTON, University of Missouri. Cholinesterase activity in the gills of several teleosts. (10 min.)

2:55 44. ALVARADO, RONALD H. and L. B. KIRSCHNER, Washington State University. Factors influencing sodium balance in the tiger salamander, *Ambystoma tigrinum.* (15 min.)

3:15 45. KAMEMOTO, FRED I. and SHARON M. KEISTER, University of Missouri. Cholinesterase activity in the crayfish kidney. (15 min.)

3:35 46. Paper cancelled. Abstract published anyway.
3:55 47. WINKERT, JOHN W., ALBERT S. GORDON and ELIZABETH L. WINKERT, New York University and New York Medical College. Reticulocyte release patterns following erythropoietic stimulation. (15 min.)
4:15 48. BENOLKEN, R. M., University of Minnesota. Recovery processes in the eye. (15 min.)

Section F. *Invertebrate Zoology* 2:00 P.M.

Lilly Life Sciences Building, Room G-431

BERTIL G. ANDERSON, *presiding*

2:00 49. LANE CHARLES E., University of Miami. Observations on the general biology of Physalia. (15 min.)
2:20 50. OGREN, ROBERT E., Dickinson College. A contribution to the biology of the land planarian, *Microplana atrocyaneus* (Walton, 1912). (10 min.)
2:35 51. KATZ, FRANK F., Jefferson Medical College. The distribution of the nematode *Strongyloides ratti* in the small intestine of gonadectomized and sham operated male and female rats. (10 min.)
2:50 52. ANDERSON, JOHN MAXWELL, Cornell University. Structural peculiarities of the pyloric caeca in particle-feeding sea-star, *Porania pulvillus*. (10 min.)
3:05 53. FARMANFARMAIAN, A., University of Shiraz, Iran and University of California, Berkeley. Storage and translocation of nutrients in the purple sea urchin (*Strongylocentrotus purpuratus*). (15 min.)
3:25 54. BOVBJERG, RICHARD V., University of Iowa. Mechanisms isolating two species of crayfish. (15 min.)
3:45 55. EDGAR, ARLAN L., ALMA COLLEGE. Mating and oviposition in the phalangid *Leiobunum longipes* (Arachnoidea: Opiliones). (15 min.)

MONDAY EVENING SESSION, AUGUST 28

Business Meeting

Division of Invertebrate Zoology 7:00 P.M.

Lilly Life Sciences Building, Room G-431

Refresher Course: *Comparative Neurophysiology*, Discussion and Question Session 8:00 P.M.

Lilly Life Sciences Building, Room 1-105

The AIBS General Meeting for all Societies 9:00 P.M.
Hall of Music

TUESDAY MORNING SESSION, AUGUST 29

Symposium: *Comparative physiology of excretory organs, I.* 9:00 A.M.

Organized by Leonard B. Kirschner for the Division of Comparative Physiology of the American Society of Zoologists. Supported by a grant from the National Science Foundation. No abstracts published for this symposium.

Lilly Life Sciences Building, Room 1-105

L. B. KIRSCHNER, *presiding*

9:00 56. OKEN, DONALD E., Peter Bent Brigham Hospital, Boston. Recent studies on the vertebrate nephron. (45 min.)

10:00 57. BENTLEY, PETER, University of Bristol. The mechanism of action of hormones involved in renal function. (30 min.)

10:45 58. SCHMIDT-NIELSEN, BODIL, Duke University, Durham, N. C. Comparative physiology of water excretion. (45 min.)

Section G. *Animal Behavior and Sociobiology, III.* 9:00 A.M.
(In collaboration with the Section of Animal Behavior and Sociobiology of the Ecological Society of America)

Smith Hall, Room 108

J. C. BRADDOCK, *presiding*

9:00 59. SIEGEL, P. B. and H. L. MARKS, Virginia Polytechnic Institute. Interactions among chickens in different levels of the social hierarchy. (15 min.)

9:20 60. REESE, E. S., University of Hawaii. Inter- and intraspecific dominance relationships of sympatric species of intertidal hermit crabs. (Motion picture, 15 min.)

9:40 61. BARLOW, G. W., University of Illinois. Color patterns as indicators of motivation in the teleost fish *Badis badis.* (15 min.)

10:00 62. FICKEN, R. W., Cornell University. Sexual behavior of the Common Grackle, *Quiscalus quiscula.* (Motion picture, 15 min.)

10:20 63. CARBAUGH, B. T. and M. W. SCHEIN, Pennsylvania State University. Sexual response of roosters to full and partial models. (15 min.)

10:40 64. BROCKWAY, B. F., Cornell University. The effects of visual and vocal stimuli upon the reproductive ethology and physiology of paired Budgerigars (*Melopsittacus undulatus*). (15 min.)

11:00 65. BANKS, E. M., A. V. NALBANDOV and W. H. JOHNSON, University of Illinois. Motor components of sexual behavior in sheep. (Motion picture, 15 min.)

11:20 66. KING, J. R. and D. S. FARNER, Washington State University. The relationship of fat deposition and vernal migratory behavior as indicated by Zugunruhe in *Zonotrichia leucophrys gambelii.* (15 min.)

Section H. *Histology and Cytology* 9:00 A.M.

Lilly Life Sciences Building, Room G-126

ROBERT C. KING, *presiding*

9:00 67. MORIBER, LOUIS G., LAWRENCE PRUTKIN and MARK E. PARKS, Brooklyn College. The nature of differential staining with acid dyes. (15 min.)

9:20 68. GOODGE, WILLIAM R., West Virginia University. A histological and histochemical study of the nasal gland of some non-marine birds. (15 min.)

9:40 69. BREMILLER, RUTH ALLEN, HISAKO O. YOKOYAMA and NAOMI LEMKEY, Medical School, Northwestern University. Phosphomonoesterases of the rat duodenum and heart. (15 min.)

10:00 70. BIRGE, WESLEY J., University of Illinois. Cytoplasmic RNA in the parenchymal liver cells of the hibernating ground squirrel (*Citellus tridecemlineatus*). (15 min.)

10:20 71. WALKER, ROWLAND, Rensselaer Polytechnic Institute. Fine structure of a virus tumor of fish. (15 min.)

10:40 72. SCHMIDT, ANTHONY J., College of Medicine, University of Illinois. Architectural regularity of the subepidermal reticulum of the adult newt, *Triturus viridescens*. (15 min).

11:00 73. KESSEL, RICHARD G. and NORMAN E. KEMP, Marine Biological Laboratory, Bowman Gray School of Medicine and University of Michigan. Electron microscope observations on the follicular envelope and developing oocyte in the tunicate, *Molgula manhattensis*. (15 min.)

11:20 74. BEAMS, H. W. and EVERETT ANDERSON, State University of Iowa. Fine structure of "intracellular ductules" in certain glands of the carabid beetle. (10 min.)

11:35 75. BAKER, CLINTON L., Southwestern College. Spermatozoa of *Amphiuma tridactylum*: morphology, helical motility and reversibility. (Motion picture, 15 min.)

Section I. *Experimental Biology* 9:00 A.M.

Lilly Life Sciences Building, Room G-431

RICHARD C. SANBORN, *presiding*

9:00 76. SAEKI, YUICHI and KEITH I. BROWN, Ohio Agricultural Experiment Station, Wooster, Ohio. Comparison of fertilizing ability of ejaculated and vasa deferentia spermatozoa in turkeys. (15 min.)

9:20 77. NASH, DONALD J. and JOHN W. GOWEN, Pennsylvania State University and Iowa State University. Reproductive performance of mice exposed to embryonic X-irradiation. (15 min.)

9:40 78. KIDWELL, J. F., D. G. NASH, JANICE STADLER and J. W. GOWEN, Iowa State University. An X-ray induced deficiency in linkage group VI of the mouse. (15 min.)

10:00 79. DOWNS, WILLIAM G., JR. and DAVID FITZPATRICK, Tennessee Polytechnic Institute, Cookeville, Tennessee. Behavior of leucocytes and serum-proteins as a result of whole-body irradiation with large dosages of gamma rays. (15 min.)

10:20 80. MONESI, VALERIO, Biology Division, Oak Ridge National Laboratory and Biology Division of the National Committee for Nuclear Energy, Research Group for Embryology, Frascati (Roma), Italy. Study of DNA synthesis and the mitotic cycle in spermatogonia of the mouse, using tritium-labeled thymidine. (Introduced by E. F. Oakberg) (15 min.)

10:40 81. SHAW, DON H. and J. R. PATEL, University of Wisconsin. Species differences between the American bison and domestic cattle as determined by heteroimmune hemolysins. (Introduced by Seymour Abrahamson) (15 min.)

11:00 82. BOND, H. E., Biology Division, Oak Ridge National Laboratory. A sex-associated protein in liver tissues of the rat and its response to castration and hormone treatment. (Introduced by Norman G. Anderson) (15 min.)

11:20 83. SHERMAN, J. K., University of Arkansas School of Medicine. Functional survival of cells during formation and dissolution of intracellular ice. (15 min.)

Section J. *Developmental Biology, I.* 9:00 A.M.

Jointly sponsored by the Division of Developmental Biology of the American Society of Zoologists and the Society for the Study of Development and Growth.

Biochemistry, Room 105

JOHN M. CAIRNS, *presiding*

9:00 84. WESSELS, NORMAN K., Stanford University. Interactions of dermis and epidermis during embryogenesis. (15 min.)

9:20 85. GRABOWSKI, CASIMER T., University of Miami. Studies on the development of the notochord-forming center of the chick embryo. (15 min.)

9:40 86. REISS, SHIRLEY E., Washington University. Growth control of neurons in the superior cervical ganglion of the mouse. (Introduced by Viktor Hamburger) (15 min.)

10:00 87. REAMS, WILLIE M., JR. and THOMAS C. MAYER, Louisiana State University. The occurrence and migration of pigment cells in the hind limb musculature of PET mice. (15 min.)

10:20 88. QUEVEDO, WALTER C., JR., JOHN M. CAIRNS and JEAN A. SMITH, Brown University and Roswell Park Memorial Institute. The effects of croton oil and 7,12-dimethylbenz(*a*)anthracene (DMBA) on melanocytes in hamsters. (15 min.)

10:40 89. RAFFERTY, NANCY S., Medical School, Johns Hopkins University. Homologous implantation of embryos into adult frogs. (15 min.)

11:00 90. PIERRO, LOUIS J., University of Connecticut. Teratogenic action of actinomycin D. (Introduced by Walter Landauer) (15 min.)

11:20 91. DELANNEY, LOUIS E., Wabash College. Homografting of sexually

mature Mexican axolotls, *Siredon mexicanum*. (15 min.)

11:40 92. VAN ALTEN, PIERSON J., College of Medicine, University of Illinois. Modification of graft-versus-host reaction in the chick embryo. (15 min.)

12:00 93. BARCH, STEPHANIE H., Michigan State University. Serological analysis of Lucké renal adenocarcinoma. (15 min.)

Lunch: Annual Luncheon, Division of Animal Behavior and Sociobiology
12:00 NOON The Union, Room 250

TUESDAY AFTERNOON SESSION, AUGUST 29

Symposium: *Comparative physiology of excretory organs, II.* 1:30 P.M.

Lilly Life Sciences Building, Room 1-105

A. W. MARTIN, *presiding*

1:30 94. CRAIG, RODERICK, University of California, Berkeley. Urine formation by the insect malpighian tubule. (45 min.)

2:30 95. RIEGEL, J. A., Queens University, Canada. Urine formation by the crustacean antennal gland. (45 min.)

3:30 96. MARTIN, A. W., University of Washington. Urine formation by the molluscan kidney. (45 min.)

Section K. *Animal Behavior and Sociobiology, IV.* 2:00 P.M.
(In collaboration with the Section of Animal Behavior and Sociobiology of the Ecological Society of America)

Smith Hall, Room 108

W. C. DILGER, *presiding*

2:00 97. BLAUVELT, HELEN, LEONARD HERSHER and A. ULRIC MOORE, New York State College of Medicine and Cornell University. The social behavior of goats deprived of maternal and herd care. (15 min.)

2:20 98. EMLEN, J. T., University of Wisconsin. Cliff edge responses of gull chicks. (15 min.)

2:40 99. DILGER, W. C., Laboratory of Ornithology, Cornell University. Changes in nest-material carrying behavior of F_1 hybrids between *Agapornis roseicollis* and *A. personata fischeri* during three years. (15 min.)

3:00 100. KLOPMAN, R. B., Cornell University. Nest building and reproductive behavior in *Agapornis* parrots. (15 min.)

3:20 101. COLLIAS, N. E., P. J. FRUMKES, D. S. BROOKS and R. J. BARFIELD, University of California, Los Angeles, Nest building and breeding behavior by castrated village weaverbirds (*Textor cucullatus*). (15 min.)

3:40 102. TOBACH, E., T. C. SCHNEIRLA, R. LAUPHEIMER and K. GROSSMAN, American Museum of Natural History. The use of a computer in a study of the behavior of rats during the litter period. (15 min.)

Section L. *Comparative Physiology, III.* 2:00 P.M.

Lilly Life Sciences Building, Room G-126

KENNNETH D. ROEDER, *presiding*

2:00 103. DUBBS, CARBON C. and RICHARD C. SANBORN, Purdue University. The osmotic pressure of the blood of individual insects. (15 min.)
2:20 104. BROOKES, VICTOR J., Oregon State University. Metabolism of the insect fat body. (15 min.)
2:40 105. MELLON, DEFOREST, JR. and DAVID R. EVANS, Johns Hopkins University. Electrophysiological evidence that water stimulates a fourth sensory cell in the blowfly taste receptor. (15 min.)
3:00 106. ROEDER, KENNETH D., Tufts University. Ultrasonic echoes from flying moths. (Motion picture, 15 min.)
3:20 107. JONES, JACK COLVARD, University of Maryland. Observations on sexually depleted male *Aedes aegypti* (L). (5 min.)

Section M. *Comparative Endocrinology, I.* 2:00 P.M.

Lilly Life Sciences Building, Room G-431

S. A. MATTHEWS, *presiding*

2:00 108. WALL, BETTY J. and C. L. RALPH, University of Pittsburgh. A histological study of cockroach neurosecretory cells following dehydration. (Introduced by Ray L. Watterson) (15 min.)
2:20 109. RALPH, C. L., University of Pittsburgh. Distribution of cardio-accelerator hormone in *Periplaneta americana.* (15 min.)
2:40 110. ROUNDS, HARRY D. and MARLENE MCCLAIN, University of Pittsburgh. Segmental distribution and response of insect chromatophorotropins. (Introduced by Paul A. Wright) (15 min.)
3:00 111. BAGNARA, JOSEPH T., University of Arizona. Onset of pineal and hypophyseal regulation of melanophores in *Xenopus.* (15 min.)
3:20 112. TEAGUE, ROBERT S. and JANE R. PATTON, University of Alabama Medical Center. An *in vivo* assay of melanophore hormone. (15 min.)
3:40 113. Paper cancelled. Abstract printed anyway.

Section N. *Developmental Biology, II.* 2:00 P.M.

Jointly sponsored by the Division of Developmental Biology of the American Society of Zoologists and the Society for the Study of Development and Growth.

Biochemistry, Room 105

ROBERT AUERBACH, *presiding*

2:00 114. HEIM, WERNER G., Wayne State University. A reproduction-associated protein in the rat. (15 min.)

2:20 115. WEINERTH, JOHN L., CHARLES R. BATTAGLIA and HULDA MAGALHAES, Bucknell University. The non-essential nature of seminal vesicles in golden hamsters. (15 min.)

2:40 116. MALONE, THOMAS E. and K. KENNETH HISAOKA, Loyola University, Chicago. The formation of deutoplasmic components in the zebrafish ovary. (15 min.)

3:00 117. NORMAN, CHARLES, C. E. JOHNSON, I. D. PORTERFIELD, E. GOLDBERG and R. S. DUNBAR, JR., West Virginia University. Prolonged maintenance of mammalian sperm in a chemically defined medium at room temperatures. (15 min.)

3:20 118. SHIVERS, ALEX, Michigan State University. Studies of the effect of homologous and heterologous anti-jelly sera on fertilization of eggs of *Rana pipiens*. (Introduced by John R. Shaver) (15 min.)

3:40 119. TYLER, ALBERT, ALBERTA SEATON and JACQUES SIGNORET, California Institute of Technology. Further analysis of anti-developmental action of antisera against fertilizins and against other egg-extracts of sea urchins. (15 min.)

4:00 120. KOCH, ELIZABETH A. and R. C. KING, Northwestern University. The origin of the vitelline membrane of *Drosophila melanogaster*. (15 min.)

4:20 121. MAHOWALD, ANTHONY P., Johns Hopkins University. Electron microscopy of early embryogenesis in *Drosophila melanogaster*. (Introduced by T. R. F. Wright) (15 min.)

4:40 122. SEVERSMITH, HERBERT F., American University. Cleavage in the brook lamprey, *Lampetra aepyptera*. (15 min.) (Abstract withdrawn)

Tea: *Division of Invertebrate Zoology* 4:00 P.M.

The Union, Room 240

Interim Business Meeting: *American Society of Zoologists* 5:15 P.M.

Lilly Life Sciences Building, Room 1-105

TUESDAY EVENING SESSION, AUGUST 29

Business Meeting: *Division of Comparative Physiology* 8:00 P.M.

Lilly Life Sciences Building, Room G-126

Section O. *Developmental Biology, III.* 7:30 P.M.

Jointly sponsored by the Division of Developmental Biology of the American Society of Zoologists and the Society for the Study of Development and Growth.

Lilly Life Sciences Building, Room 1-105

THOMAS J. KING, *presiding*

7:30 123. LEMKEY, NAOMI and HISAKO O. YOKOYAMA, Medical School, Northwestern University. Esterases and phosphatases of the developing and adult mouse brain. (15 min.)

7:50 124. ALLEN, SALLY LYMAN, University of Michigan. The role of the esterase isozymes in *Tetrahymena*. (15 min.)

8:10 125. NUNNALLY, DAVID A., Vanderbilt University. Development of succinic dehydrogenase in the duodenum of the chick. (Introduced by Florence Moog) (15 min.)

8:30 126. COLEMAN, JOHN R., Carnegie Institution of Washington and Johns Hopkins University. Deoxyribonucleases in embryonic and adult *Rana pipiens*. (Introduced by James D. Ebert) (15 min.)

8:50 127. FRASER, RONALD C., University of Tennessee. The development of hemoglobins in the chick. (15 min.)

9:10 128. KOSIN, I. L. and S. H. WEISBROTH, Washington State University. "Physiological zero" of turkey blastoderms and genetic differences in their mitotic index. (15 min.)

9:30 129. DIBERARDINO, MARIE A., Institute for Cancer Research. The karyotype of *Rana pipiens* and investigation of its stability during embryonic differentiation. (Introduced by T. J. King) (15 min.)

9:50 130. VOLPE, E. PETER and SANJOY DASGUPTA, Newcomb College of Tulane University. Development, sex and pigmentary patterns of gynogenetic diploid leopard frogs *(Rana pipiens)*. (15 min.)

10:10 131. SHELL, LESTER C., University of Dubuque. Comparison of mitotic figure distribution in the central nervous system of normal and colchicine-treated *Rana pipiens* embryos. (15 min.)

WEDNESDAY MORNING SESSION, AUGUST 30

Symposium: *Origin, structure and functioning of hemocytes in invertebrates, I.* 9:00 A.M.

Organized for the Division of Invertebrate Zoology of the American Society of Zoologists by Melbourne R. Carriker.

Biochemistry, Room 105

JOHN M. ANDERSON, *presiding*

9:00 132. TONEY, MARCELLUS E., JR., Virginia Union University. The morphology and origin of blood cells of several Crustacea.

9:40 133. JONES, JACK COLVARD, University of Maryland. Current concepts concerning insect hemocytes.

10:20 134. RIZKI, M. T. M., Reed College. Experimental analysis of hemocyte morphology.

11:00 135. WITTIG, GERTRAUDE, U. S. Department of Agriculture, Beltsville, Maryland. Investigations of blood cells in insect pathology.

Section P. *Developmental Biology, IV.* 9:00 A.M.

Jointly sponsored by the Society for the Study of Development and Growth, the Developmental Biology Section of the Botanical Society of America and the Division of Developmental Biology of the American Society of Zoologists.

Lilly Life Sciences Building, Room 1-105

JOHN W. SAUNDERS, JR., *presiding*

9:00 136. JACOBS, WILLIAM P. and CLIFFORD E. LA MOTTE, Princeton University. Physiological investigations of phloem regeneration and its regulation in *Coleus* internodes. (15 min.) (No abstract)

9:20 137. ROBERTS, LORIN W., University of Idaho. Experimental modification of the pattern of wound-xylem differentiation in *Coleus* stem wound-meristems. (15 min.) (No abstract)

9:40 138. LASH, J., F. ZILLIKEN and F. A. HOMMES, Medical School, University of Pennsylvania and R. K. Universiteit, The Netherlands. Chondrogenic induction with a low molecular weight tissue component. (15 min.)

10:00 139. CLUTTER, MARY E. and IAN M. SUSSEX, Yale University. Chemically induced differentiation in sterile-cultured tobacco pith. (15 min.) (No abstract)

10:20 140. STOCKDALE, FRANK, JOAN ABBOTT and HOWARD HOLTZER, Medical School, University of Pennsylvania. Behavior of cartilage cells *in vitro*. (15 min.)

10:40 141. AUERBACH, ROBERT, University of Wisconsin. Effects of transplantation on thymus lymphoid differentiation. (15 min.)

11:00 142. NEGLIC, MOSHE and ANTON LANG, California Institute of Technology. Studies on orientation of cell division in *Hyoscyamus niger*. (15 min.) (No abstract)

11:20 143. WHITE, RICHARD H., Washington University. An analysis of the development of the compound eye in the mosquito, *Aedes aegypti*. (15 min.)

11:40 144. HABER, ALAN H. and WILLIAM L. CARRIER, Oak Ridge National Laboratory. Nonessentiality of cell division for leaf morphogenesis during growth of young wheat seedlings. (15 min.) (No abstract)

Section Q. *Behavioral Films, I. Animal Behavior and Sociobiology, V.*
9:00 A.M.

(Cosponsored by the Section of Animal Behavior and Sociobiology of the Ecological Society of America; no abstracts)

Smith Hall, Room 108

E. M. BANKS, *presiding*

9:00 145. CARPENTER, C. C. and KIRK J. MOSLEY, III, University of Oklahoma. Patterns of display in Iguanid lizards. (30 min.)

9:35 146. COLLIAS, N. E., University of California, Los Angeles. Breeding behavior of the Blackheaded Weaverbird (*Textor cucullatus*). (15 min).

9:55 147. BLAUVELT, H. and A. U. MOORE, New York State College of Medicine, Syracuse. Population-destructive intraspecies fighting. (15 min.)

10:15 148. GINSBURG, B. E., G. B. RABB, and S. L. ANDREWS, University of Chicago. Comparative studies of Canid behavior. I. Birth and maternal behavior. (20 min.)

10:45 149. GINSBURG, B. E., G. B. RABB and S. L. ANDREWS, University of Chicago. Comparative studies of Canid behavior. II. Development of social behavior in relation to early experience. (25 min.)

11:15 150. BUECHNER, HELMUT E., Washington State University. Territorial behavior in Uganda Kob. (25 min.)

Section R. *Comparative Physiology, IV.* 9:00 A.M.

Lilly Life Sciences Building, Room G-126

RODERICK CRAIG, *presiding*

9:00 151. MURRAY, DARREL and EARL SEGAL, Purdue University and Rice University. The effects of temperature and light on locomotor activity of the American cockroach, *Periplaneta americana.* (15 min.)

9:20 152. MUTCHMOR, JOHN A. and A. GLENN RICHARDS, University of Minnesota. Low temperature tolerance of insects in relation to the influence of temperature on muscle apyrase activity. (15 min.)

9:40 153. ROBERTS, JOHN L., University of Massachusetts. Day to night respiration of carp, *Carassius carassius* (L), following thermal acclimation and photoperiod control. (15 min.)

10:00 154. EVANS, R. M., R. C. PURDIE, and C. P. HICKMAN, JR., University of Alberta. The effect of different temperature and photoperiod conditions on the respiratory metabolism of rainbow trout (*Salmo gairdneri*). (15 min.)

10:20 155. LICHT, PAUL, University of Michigan. Effects of temperature on heart rates of lizards during rest and activity. (Introduced by W. R. Dawson). (15 min.)

10:40 156. LASIEWSKI, ROBERT C., University of Michigan. Energy metabolism of hummingbirds. (Introduced by J. M. Allen) (15 min.)

11:00 157. FREHN, JOHN and ADAM ANTHONY, Pennsylvania State University. Respiration and phosphorylation of liver mitochondria from cold-exposed rats and chipmunks. (15 min.)

Section S. *Comparative Endocrinology, II.* 9:00 A.M.

Lilly Life Sciences Building, Room G-431

RICHARD A. EDGREN, *presiding*

9:00 158. SIEGEL, H. S. and P. B. SIEGEL, Virginia Polytechnic Institute. Genetic differences in response of chick testes to gonadotropins. (Introduced by P. C. Holt) (15 min.)

9:20 159. BRENEMAN, W. R. and FRANK J. ZELLER, Indiana University. P^{32} uptake by chick testes as an end-point for gonadotropin assay. (15 min.)

9:40 160. FOREMAN, DARHL, Western Reserve University. Some short-term effects of gonadotrophins on the metabolism of the gonads. (15 min.)

10:00 161. YOCHIM, J. M. and V. J. DE FEO, University of Illinois. Control of the decidual cell response by hormones of the ovary. (15 min.)

10:20 162. GILCHRIST, RICHARD D. and ROGER A. HOFFMAN, U. S. Army Chemical Research and Development Laboratories, Maryland. Changes in ascorbic acid and cholesterol in the cryptorchid rat testis. (Introduced by Ronald R. Novales) (15 min.)

10:40 163. VANDENBERGH, JOHN G., Pennsylvania State University. Birefringence and sudanophilia in starling and red-wing blackbird testes. (10 min.)

10:55 164. MERKLIN, ROBERT J., Jefferson Medical College. Ovarian, uterine and adrenal lipids of lactating mice. (15 min.)

11:15 165. KITAY, JULIAN I., University of Virginia School of Medicine. Sex differences in the effects of gonadectomy on pituitary and adrenal function in rats. (15 min.)

11:35 166. BERNARD, G. R., Medical College of Georgia. Some alterations of the adrenal glands of *Mesocricetus auratus* infected with *Trichinella spiralis*. (12 min.)

Lunch: *Division of Developmental Biology* 12:30 P.M.
The Union, North Ballroom

WEDNESDAY AFTERNOON SESSION, AUGUST 30

Symposium: *Origin, structure and functioning of hemocytes in invertebrates, II.* 2:00 P.M.

Biochemistry, Room 105

MELBOURNE R. CARRIKER, *presiding*

2:00 167. STAUBER, LESLIE A., Rutgers, The State University. *In vivo* studies of the activity of the blood cells of oysters. (No abstract)

2:40 168. MACKIN, J. G., Agricultural and Mechanical College of Texas. Oyster leucocytes in infectious disease.

3:20 169. BOOLOOTIAN, RICHARD A., University of California, Los Angeles. Coelomocytes of echinoderms.

4:00 170. ANDREW, WARREN, Indiana University. A study of living blood of some species of Bermuda tunicates, as observed by phase microscopy.

Section T. *Developmental Biology, V.* 2:00 P.M.

Jointly sponsored by the Society for the Study of Development and Growth, the Developmental Biology Section of the Botanical Society of America and the Division of Developmental Biology of the American Society of Zoologists.

Lilly Life Sciences Building, Room 1-105

LAWRENCE J. CROCKETT, *presiding*

2:00 171. KONIJN, THEODORE M. and KENNETH B. RAPER, University of Wisconsin. Cell aggregation in *Dictyostelium discoideum*. (15 min.) (No abstract)

2:20 172. TAKEUCHI, IKUO, Carnegie Institute of Washington. Immunochemical and histochemical studies on the development of the cellular slime mold, *Dictyostelium mucoroides*. (15 min.) (No abstract)

2:40 173. HOHL, HANS-RUDLPH, University of Wisconsin. Plastids and tumorgenesis. (15 min.) (No abstract)

3:00 174. HAGEN, GEORGE L., Institute for Cancer Research, Philadelphia. Developmental sequence of leaf tumor formation in a tobacco hybrid. (15 min.) (No abstract)

3:20 175. BRIGGS, R., J. SIGNORET and R. R. HUMPHREY, Indiana University. Transplantation of genetically marked nuclei in the axolotl. (15 min.)

3:40 176. MCKINNELL, ROBERT G., Institute for Cancer Research, Philadelphia. Intraspecific nuclear transplantation in *Rana pipiens*. (15 min.)

4:00 177. POLLOCK, EDWARD and WILLIAM A. JENSEN, University of California, Berkeley. A cytochemical analysis of embryogenesis in higher plants. (15 min.) (No abstract)

4:20 178. MAKSYMOWYCH, ROMAN, Villanova University. Cell division in leaf development of *Xanthium pennsylvanicum*, Wallr. (15 min.) (No abstract)

4:40 179. FOARD, DONALD E. and ALAN H. HABER, Oak Ridge National Laboratory. Formation of guard cells in wheat leaves during growth without cell division. (15 min.) (No abstract)

Section U. *Behavioral Films, II. Animal Behavior and Sociobiology, VI.*

2:00 P.M.

(Cosponsored by the Section of Animal Behavior and Sociobiology of the Ecological Society of America)

Smith Hall, Room 108

E. M. BANKS, *presiding*

2:00 180. Social Insects: The Honeybee. Encyclopedia Britannica Films in collaboration with Dr. A. E. Emerson.

2:35 181. The Encyclopoedia Cinematographica. Institut für den Wissenschaftlichen Film.

2:50 182. Paarungsbiologie der Anuran. Institut für den Wissenschaftlichen Film, K. Lorenz & I. Eibl-Eibesfeldt. (11 min.)

3:05 183. Sexualverhalten und Erablage beim Alpenmolch. Institut für den Wissenschaftlichen Film, K. Lorenz & I. Eibl-Eibesfeldt. (6 min.)

3:15 184. DE VORE, I. and S. L. WASHBURN, University of California, Berkeley. Baboon behavior. (31 min.)

Section V. *Demonstration Film* 4:00 P.M.

Smith Hall, Room 108

4:00 185. ELIAS, HANS, Chicago Medical School. Looking into space: a motion picture on three-dimensional interpretation of flat sections. (40 min.)

Section W. *Comparative Endocrinology, III.* 2:00 P.M.

Lilly Life Sciences Building, Room G-431

AUBREY GORBMAN, *presiding*

2:00 186 GROSSO, LEONARD L., College of Saint Teresa. Peroxidase inhibition in fetal mouse thyroid. (10 min.)

2:15 187. MULVEY, PHILIP F., JR. and D. WARD SLINGERLAND, V. A. Hospital, Boston. Stimulation of the hexose monophosphate shunt in rat thyroid slices by small amounts of propylthiouracil. (15 min.)

2:35 188. MEISNER, H. M., University of Alberta. The effect of propylthiouracil on oxygen consumption and radioiodine metabolism of the frog, *Rana pipiens*, in winter. (Introduced by C. P. Hickman, Jr.) (15 min.)

2:55 189. HARCLERODE, JACK E. and ADAM ANTHONY, Pennsylvania State University. Thyroidal uptake and turnover of I^{131} by rats exposed to simulated altitude. (15 min.)

3:15 190. ELEFTHERIOU, B. E. and M. X. ZARROW, Purdue University. The influence of thyroidectomy and thyroxine on brain maturation in two subspecies of *Peromyscus maniculatus*. (15 min.)

3:35 191. SUDDICK, RICHARD P. and THEODORE J. URBAN, Creighton University. A change in acidophilic cells of the pituitary gland of the rat after salivariadenectomy. (15 min.)

3:55 192 JOHNSON, NORMA, Marquette University. Toxic effects of adrenal grafts to the chorioallantois. (Introduced by J. W. Saunders, Jr.) (15 min.)

Business Meeting: *Division of Comparative Endocrinology* 4:30 P.M.

Lilly Life Sciences Building, Room G-431

WEDNESDAY EVENING SESSION, AUGUST 30

Zoologists' Dinner 6:30 P.M.

Purdue Union, The North Ballroom

Presidential Address, "The Challenge and Responsibility of Zoology"
by C. LADD PROSSER

PAPERS READ BY TITLE

Papers read by title have been arranged alphabetically by the name of the first author.

193. ALLEN, M. JEAN, Wilson College. Histochemical observations on the early developmental stages of the polychaete, *Arenicola claparedii.*
194. ALLEN, M. JEAN, Wilson College. Histochemical observations on the early developmental stages of the polychaete, *Autolytus ornatus.*
195. ALLEN, RUSSELL F. and JOHN F. LHOTKA, JR., University of Oklahoma Medical School. Studies on *Crotalus atrox.* III. Effects of stress on the fundic mucosa.
196. AUCLAIR, WALTER, George Washington University School of Medicine. Monolayer cultures of *Rana pipiens* kidney cells.
197. BAIRD, DONALD, Princeton University. A haplolepid fish fauna in the Early Pennsylvanian of Nova Scotia.
198. BERNARD, G. R. and J. M. WINNER, Medical College of Georgia and University of Notre Dame. Some alterations of adrenal glands of *Mesocricetus auratus* during the course of infection with the Khartoum strain of *Leishmania donovani.*
199. BIRGE, WESLEY J., University of Illinois. A cytochemical study of the differentiating ependyma in the chick embryo.
200. BOATMAN, JOSEPH B., PATRICIA A. LASKEY and MARVIN J. RABINOVITZ, William H. Singer Memorial Research Laboratory of the Allegheny General Hospital, Pittsburgh. Relation of time and mass of incubated thyroid slices to I^{131} and QO_2 metabolism.
201. BRELAND, OSMOND P. and JOHN G. RIEMANN, University of Texas. Preliminary studies of spermatogenesis in the mosquito, *Culiseta inornata* Williston.
202. CALVENTI, IDELISA and ROSS F. NIGRELLI, New York Aquarium, New York Zoological Society. Effects of copper salts on *Fundulus heteroclitus.*
203. CAMOUGIS, GEORGE, LOUIS P. GRANATH and RICHARD G. BESCHLE, Clark University and Worcester Polytechnic Institute. Preliminary studies on the navigational ability of electric fishes.
204. CHAVIN, WALTER and MADELEINE OLIVEREAU, Wayne State University and Institut Océanographique. Adrenal histochemistry of fresh water and marine teleosts.
205. DE GENNARO, LOUIS D., LeMoyne College, Syracuse. Chorioallantoic grafting of radioactive chick glycogen body.
206. DORNFEST, BURTON S., JOSEPH LOBUE, EUGENE S. HANDLER, ALBERT S. GORDON and HENRY QUASTLER, New York University and Brookhaven National Laboratory. Further studies on leukocyte release from perfused isolated femurs of rats.
207. DOUGHERTY, ELLSWORTH C., D. JOANNE FERRAL and BJORN SOLBERG, University of California, Berkeley. Xenic cultivation of antarctic micrometazoa. 1. The tardigrade *Hypsibius articus* (Murray, '07) Marcus, '28.
208. DOUGHERTY, ELLSWORTH C., D. JOANNE FERRAL and BJORN SOLBERG, University of California, Berkeley. Xenic cultivation of antarctic micrometazoa. 2. The rotifer *Philodina gregaria* Murray, '10.
209. EAKIN, RICHARD M. and JANE A. WESTFALL, University of California, Berkeley. Photoreceptors in the stirnorgan of tadpoles of the treefrog, *Hyla regilla.*

210. EVANS, LLEWELLYN T., Research Laboratory, Jaffrey Center, New Hampshire. Aquatic courtship of the wood turtle, *Clemmys insculpta.*
211. EVANS, LLEWELLYN T., Research Laboratory, Jaffrey Center, New Hampshire. An interpretation of turkey courtship.
212. FRENCH, W. L. and J. B. KITZMILLER, University of Illinois. Cytological evidence for crossing-over in males of *Anopheles quadrimaculatus.*
213. GARDNER, PAUL J. and IRWIN L. BAIRD, Omaha University and University of Kansas. Some observations on venous flow in the turtle, *Pseudemys scripta.*
214. GRANT, WILLIAM C., JR. and ALLYN J. WATERMAN, Williams College. Effect of light on thyroid activity in *Diemictylus viridescens.*
215. GROSSO, LEONARD L., College of Saint Teresa. Goitrogen-induced fetal liver alterations.
216. HEIM, WERNER G., Wayne State University. Quantitative relations between the fetal, neonatal and maternal serum proteins of the rat.
217. HERSHBERGER, L. G., C. R. THOMPSON and P. M. ZAMBOANGA, Cutter Laboratories, Berkeley, California. Effect of carrageenin on cellulose sponge assay for anti-inflammatory substances.
218. HISAOKA, K. K. and C. F. FIRLIT, Loyola University, Chicago. The distribution and localization of ribonucleic acid in the zebrafish embryo as determined by histochemical methods.
219. HOGUE, MARY J., Medical School, University of Pennsylvania. The effect of poliomyelitis on guinea pig brain cells in tissue culture.
220. HORNER, B. ELIZABETH, Smith College. Paternal care of the young and convulsive seizures in the grasshopper mouse.
221. HUDDLESTON, CARLA J. and LAWRENCE I. GILBERT, Northwestern University. Acid phosphatase activity in the testis of the Cynthia silkworm.
222. HUMPHREY, R. R., Indiana University. A chromosomal deletion in the Mexican axolotl (*Siredon mexicanum*) involving the nucleolar organizer and the gene for dark color.
223. IVES, PHILIP T., Amherst College. Some effects of Cobalt-60γ radiation on premeiotic spermatogenic cells in *Drosophila melanogaster.*
224. JAKOWSKA, SOPHIE, R. F. NIGRELLI and A. H. SPARROW, New York Aquarium and Brookhaven National Laboratory. Effects of chronic gamma irradiation on adult newts, *Diemictylus viridescens.*
225. KALLMAN, KLAUS D., New York Zoological Society. Detection of gynogenesis in the viviparous teleost, *Mollienesia formosa* (Girard), by tissue transplantation.
226. KALTER, H., Children's Hospital Research Foundation, Cincinnati. The teratogenic action of prednisolone on a genotype resistant to cortisone.
227. KAPLAN, HAROLD M. and D. J. O'BRIEN, Southern Illinois University. A comparative evaluation of the anesthetic properties of six barbiturates and of Viadril in the rabbit.
228. KESSEL, RICHARD G., Marine Biological Laboratory and Bowman Gray School of Medicine. Specialization of the free surface of ectoderm cells in planula larvae of *Aurelia aurita.*
229. KITZMILLER, J. B. and W. L. FRENCH, University of Illinois. Chromosomes of *Anopheles quadrimaculatus.*
230. KLEINHOLZ, L. H., H. ESPER, C. JOHNSON and F. KIMBALL, Reed College and Marine Biological Laboratory, Woods Hole, Mass. Characterization and partial purification of crustacean eyestalk hormones.
231. LATIMER, HOMER B., PAUL GIBBONS ROOFE and LILY S. FENG, University of Kansas and Jackson Hole Biological Station. Weights and measurements of the body and of some of the organs of the tiger salamander.

232. LEA, MALCOLM S. and LAWRENCE I. GILBERT, Northwestern University. Cell division in diapausing silkworm pupae.
233. LHOTKA, JOHN F., JR. and H. A. DAVENPORT, University of Oklahoma and Northwestern University Medical Schools. Congenital polycystic kidney in the black hooded rat.
234. LINDEMAN, V. F., Syracuse University. Phosphatases in the rat testis following treatment with Diethylstilbestrol.
235. LINDEMAN, V. F. and FREDERIC GREENBERG, Syracuse University. The effect of feeding Furadroxyl upon the 5′nucleotidase activity of the rat testis.
236. LUCE, WILBUR M., University of Illinois. The effect of lactamide upon the facet number in the mutants forked Bar, forked double Bar, forked Infrabar-Bar and forked wildtype in *Drosophila melanogaster*.
237. MAY, RAOUL MICHEL and FRANCOISE LAHILLE, Faculté des Sciences, University of Paris. Direct action of X-rays on the spleen of chick embryos during its myelogenesis.
238. METCALF, J. F. and J. R. FISHER, Florida State University. Glutaminase activity in the brain of developing chick embryos.
239. MORRILL, JOHN B., Wesleyan University. Development of *Lymnaea stagnalis* eggs with giant polar bodies.
240. MORROW, JAMES E., University of Alaska. Zoogeography of *Chauliodus* (Pisces, Clupeiformes).
241. MUSACCHIA, X. J. and MELCHIOR ISTVAN CHLADEK, St. Louis University. Investigations of the cloacal bladders in turtles.
242. NAGABHUSHANAM, R., Andhra University. Response of marine borers to chemically treated timbers.
243. NAGABHUSHANAM, R., Andhra University. Ecological factors related to the distribution of *Bankia campanellata* in Visakhapatnam Harbour.
244. NORMAN, CHARLES, J. L. MCCOY, I. D. PORTERFIELD and H. A. WILSON, West Virginia University. Elimination of *Vibrio fetus* from bovine sperm stored at low temperatures.
245. PARKS, MARK E. and LOUIS G. MORIBER, Brooklyn College. A freezing-substitution method permitting the cytochemical demonstration of lipids.
246. PIETSCH, PAUL, Bowman Gray School of Medicine and Wake Forest College. Effects of X-rays on regenerating limb and tail blastemata.
247. PIKÓ, LAJOS, California Institute of Technology. *In vitro* observation of cleavage of dispermic rat eggs.
248. PIKÓ, LAJOS, California Institute of Technology. A study on the possibility of dispermic androgenesis in X-irradiated rat eggs.
249. RAMM, GORDON M., University of Maryland. Development of chick embryos under low level gravitational stress.
250. RUBEN, LAURENS N. and JEAN STEVENS, Reed College. Some aspects of homograft-induced accessory limbs in urodeles. I. Systemic dosage effects.
251. RUBEN, LAURENS N. and JEAN STEVENS, Reed College. Some aspects of homograft-induced accessory limbs in urodeles. II. Local dosage effects.
252. RUGGIERI, GEORGE D., S. J., MORRIS H. BASLOW, SOPHIE JAKOWSKA and R. F. NIGRELLI, New York Aquarium, New York Zoological Society. Developmental aberrations in sea-urchin eggs induced by sponge extracts.
253. RUGH, ROBERTS, Columbia University. Sequelae to low-level X-irradiation of the mammalian embryo.
254. RULON, OLIN, Northwestern University. The extension of fertilizability and life in sand dollar eggs with vitamin B_{12}.
255. SCHECHTMAN, A. M. and CHARLES C. CHAMBERLAIN, University of California, Los Angeles. Effects of various doses of antigen on suppressed birds.

256. SCHECTMAN, A. M. and HECTOR TIMOURIAN, University of California, Los Angeles. Enhancing effect of cortisone on antibody production by chickens.
257. SEHE, CHARLES T., State University of Iowa and University of Cincinnati. Radioautographic studies on the ultimobranchial body and thyroid gland in vertebrates: reptiles and birds.
258. SIMMONS, J. EMMETT, BERTIE F. ARGYRIS and THOMAS S. ARGYRIS, Syracuse University. Effect of Ehrlich ascites tumor transplants on the mammary duct epithelium of gestating and lactating mice.
259. SINDERMANN, CARL J., U. S. Bureau of Commercial Fisheries Biological Laboratory, Boothbay Harbor, Maine. The effect of larval trematode parasites on snail migrations.
260. SINDERMANN, CARL J., U. S. Bureau of Commercial Fisheries Biological Laboratory, Boothbay Harbor, Maine. Isoagglutination in elasmobranch fishes.
261. SONNEBORN, T. M. and R. V. DIPPELL, Indiana University. The limit of multiplicity of cortical organelle systems in *Paramecium aurelia*, syngen 4.
262. SOUTHWICK, CHARLES H. and MIRZA AZHER BEG, Ohio University and Aligarh Muslim University. Social behavior of rhesus monkeys in a temple habitat in northern India.
263. SPEIDEL, CARL CASKEY, University of Virginia. Induction by ultraviolet irradiation of variations in the micronucleus of *Tetrahymena corlissi*.
264. SQUADRONI, JOSE, S. J. and ALEXANDER WOLSKY, Fordham University. Direct demonstration of sexual differences in skin homografts of the newt *Triturus viridescens*.
265. STINSON, BENJAMIN D., Princeton University. Differential effects of normal autografts of whole versus lateral half forearm regenerates on X-irradiated upper arms of adult newts.
266. SUDAK, FREDERICK N. and WALTER B. ESSMAN, Albert Einstein College of Medicine. Memory and problem solving after reanimation from deep hypothermia.
267. TYLER, WALTER S., RICHARD F. MCLAUGHLIN, JR. and ROBERT O. CANADA, University of California, Davis. Branches of the bronchial artery to the interalveolar septa of the horse lung.
268. TYLER, WALTER S., JACK PANGBORN and LOGAN M. JULIAN, University of California, Davis. Fine structure of the air capillaries of the avian lung.
269. WALTON, ARTHUR C., Knox College. Amphibia as hosts of nematode parasites. I.
270. WALTON, ARTHUR C., Knox College. Amphibia as hosts of nematode parasites. II.
271. WATERMAN, ALLYN J., Williams College. Thyroid gland of *Scyliorhinus (Scyllium) stellare*.
272. WEISS, ANDREW B., SAMUEL A. MATTHEWS and WILLIAM C. GRANT, JR., Williams College. Some effects of temperature and insulin on carbohydrate metabolism in the frog, *Rana pipiens*.
273. WILBER, CHARLES G. and PAUL F. ROBINSON, Kent State University and U. S. Army Chemical Research and Development Laboratories. Role of liver glycogen in swimming performance of white mice.
274. WOLSKY, MARIA de ISSEKUTZ, SUZAN REGAN and ALEXANDER WOLSKY, Fordham University and Manhattanville College, Purchase, New York. Differences in the facet number of Bar heterozygotes from various inbred populations of *Drosophila melanogaster*.

Abstracts

Abstracts are arranged in a single sequence alphabetically according to the last name of the first author. Papers 193 to 274 are read by title. The number of an abstract will give a clue to its location in the program. An author index follows the abstracts.

193

M. JEAN ALLEN, Wilson College. Histochemical observations on the early developmental stages of the polychaete, *Arenicola Claparedii*.

The cytoplasm and nucleolus of the flattened developing egg of *Arenicola* are strongly basophilic. The cytoplasm of fully grown eggs stains less intensely except for a cortical rim of RNA. Interphase nuclei of cleaving blastomeres are surrounded by a wide band of RNA; during mitosis RNA becomes concentrated in the amphiastral figure which stains intensely. Gastrulae to elongated trochophores (4 1/3-day larvae) have a periphero-central gradient of RNA, a consequence of the central concentration of endodermal yolk spheres lacking RNA.

DNA and basic proteins (probably histones) were demonstrated in nuclei of the cleavage and larval stages tested but appear to be too dispersed to be demonstrated in the germinal vesicle.

The cytoplasm of fully grown eggs is packed with fine glycogen granules; larger granules form a cortical rim. At maturation the residual substance stains intensely. During cleavage the cytoplasm of the blastomeres is strongly positive for glycogen, as are amphiastral figures. By the trochophore stage, glycogen is more concentrated in gut endoderm, and in 10-day larvae it is localized mainly in post-pharyngeal gut. The egg membrane (and possibly the cortex) contains a non-glycogen polysaccharide (or possibly a muco- or glyco-protein).

Lipids are present in the nucleolus of the germinal vesicle, and numerous lipid spherules are diffusely distributed throughout the cytoplasm of unfertilized eggs. During early cleavage lipid spherules are distributed to the cytoplasm of all blastomeres. By the gastrula and larval stages lipids are more concentrated in gut endoderm.

Alkaline phosphatase tests were negative for all stages tested through 10-day larvae. (Embryological stages were obtained at the Stazione Zoologica, Naples, Italy. This research was begun under a post-doctoral fellowship from the U.S.P.H.S. of the National Cancer Institute and is currently being supported by grant G-8775 from the N.S.F.)

194

M. JEAN ALLEN, Wilson College. Histochemical observations on the early developmental stages of the polychaete, *Autolytus ornatus*.

This syllid bears a ventral egg sac in which all developing eggs are at approximately the same stage. Eggs maturing in the coelom are packed with yolk spheres which are negative for RNA; as a consequence they exhibit very weak cytoplasmic basophilia, RNA being distributed in the small amount of ergastoplasm surrounding the yolk spheres. During meiosis, and during mitoses of cleaving blastomeres in the egg sac, the amphiastral figures are intensely basophilic; under oil, astral rays appear as linearly arranged rod-shaped granules of RNA. Differentiating endodermal cells have little or no cytoplasmic basophilia, while ectodermal and mesodermal cells retain their cytoplasmic basophilia through the larval stages studied.

DNA and basic proteins (probably histones) were demonstrated (method of Alfert and Geschwind, 1953) in the nuclei of coelomic eggs, cleavage blastomeres and larval stages. Strongly positive reactions occurred in the cytoplasm of mucus-secreting cells of larvae (compare results of Horn and Ward, 1957, on salivary gland cytoplasm).

Protein is present throughout most of the cell in coelomic eggs and throughout all stages tested to swimming larvae. Dense accumulations of protein occur in yolk spheres (especially), nuclear membranes, chromatin material, and also (later stages) in cell membranes, cilia, larval membranes, and, in particular, cytoplasm of mucus-secreting cells.

Alkaline phosphatase tests were reported as negative up to prehatching larvae (Allen, 1957). Tests using substrates for longer periods (18 hours) indicate that alkaline phosphatase is becoming evident in the gut of elongating trochophores (a few within one egg sac gave positive results). (Embryological stages were obtained at the Institute of Marine Biology, Parguera, Puerto Rico. This research was begun under a post-doctoral fellowship from the U.S.P.H.S. of the National Cancer Institute and is currently being supported by grant G-8775 from the N.S.F.)

195

RUSSELL F. ALLEN and JOHN F. LHOTKA, Jr., University of Oklahoma Medical School. Studies on *Crotalus atrox*. III. Effects of stress on the fundic mucosa.

The western diamondback rattlesnake is extremely sensitive to stress stimuli and death is usually inevitable if prolonged. Captivity alone may be sufficient to kill the animal (Proc. Okla. Acad. Sci. 40, 19). In mammals, stress may induce pathological alterations (gastric ulcerations, etc.) but in previous investigations (Anat. Rec. 139, 202, 292) on the normal morphology and polysaccharide histochemistry of rattlesnake fundic glands, similar changes were not observed. To study specifically the effects of stress on these glands, gross and microscopic tissue comparisons were made (processed histologically and histochemically as in the earlier work) on stressed (daily simple handling until sacrifice) and unstressed (sacrificed shortly after capture) animals. Generally no gross or microscopic tissue alterations appeared to be induced by this treatment and the three basic fundic cell types described earlier (surface columnar cells, rounded

neck cells, larger gland cells) remained unchanged. There were no pathological changes (inflammatory cells, congestion, etc.) present in the mucosa. Histochemically, unstressed animals demonstrated strong PAS neutral polysaccharide and Best carmine glycogen (diastase controlled) localizations in the columnar and neck cells. Stressed specimens showed reduced PAS staining and a complete absence of glycogen. Acid polysaccharides (Hale and Alcian blue technics) were weakly positive in the columnar cell luminal tips and were unaffected by stress. It would appear that under the limited conditions of this experiment reduction of neutral polysaccharides and glycogen depletion are the only visible effects of stress in the diamondback rattlesnake fundic mucosa in contrast to the marked changes often found in mammals. (Supported by National Institute of Health grant H-1907 (C6) and Fellowship PX-73-1.)

124

SALLY LYMAN ALLEN, University of Michigan. The role of the esterase isozymes in *Tetrahymena*. (15 min.)

In higher organisms Markert and Moller ('59) showed that an enzyme could exist in multiple form and that the activity of these isozymes varied in different species, different adult tissues and within the same tissue during ontogeny. Analysis of an isozymic series in a microorganism should provide insight into the control and role of these isozymes in the cell. Using starch gel electrophoresis, an isozymic series of esterases is currently being investigated in variety 1 of *Tetrahymena pyriformis*. A single locus controls the series. Each homozygote produces four to five isozymes at pH 7.5, while the heterozygote has the potentiality of producing all eight or nine isozymes. Clonal analyses reveal that all of the isozymes of a given genotype are present in a single cell. Variation in growth conditions results in changes in activity of individual isozymes, and under some conditions homologous changes occur between genotypes. Several cell fractionation procedures are being employed: differential centrifugation, alcohol-salt for preparing cilia (Preer and Preer '59), and digitonin-KCl for preparing pellicles and kinetosomes (Seaman '60). These studies show that one of the isozymes in each genotype is present in a small particle. The other isozymes are limited to the pellicle fraction, and one of these may differ from the others in its solubility in digitonin-KCl. These results favor the hypothesis, in part at least, that an isozyme may arise as a consequence of its site of attachment within the cell. Such attachment might permit channeling of a single enzyme to different biochemical pathways. (Supported by grant C-3545 from the National Cancer Institute, U.S.P.H.S.)

44

RONALD H. ALVARADO and L. B. KIRSCHNER, Washington State University. Factors influencing sodium balance in the tiger salamander, *Ambystoma tigrinum*. (15 min.)

Larval and adult *A. tigrinum* remain in a steady state with respect to sodium when kept in artificial pond water containing 1.2 microequivalents of sodium per ml. The concentration of sodium in the plasma of these animals is approximately 90 microequivalents per ml so that a large concentration difference exists between the internal and external environments. This concentration gradient is maintained by the active uptake of sodium across the body surface. The magnitudes of influx and outflux of sodium were estimated by use of radioactive sodium (Na^{22}). Under steady state conditions influx over the body surface balances renal and extrarenal loss of sodium, the flux values approximating 1-2 microequivalents of sodium per hour per 10 grams of body weight.

When larval *A. tigrinum* are placed in deionized distilled water and the concentration of sodium in the external bath is monitored over a long period of time (5 days) three things are observed. There is an initial rapid increase in sodium concentration in the bath, indicating a net loss of sodium on the part of the animal. After 40-45 hours the external concentration reaches a maximum and then declines, indicating a net uptake of sodium on the part of the animal. Finally the bath approaches an "equilibrium value" of about 0.05 microequivalents per ml.

It is evident from this experiment as well as from other experiments that exposure of these animals to distilled water for a sufficient period of time enhances their capacity to take up sodium. Flux studies (employing Na^{22}) revealed that two factors contribute to this process, the influx is increased and the efflux is decreased. This can be explained if one postulates the existence of an inwardly oriented, carrier type, sodium transport mechanism which is incapable of distinguishing between sodium ions derived from the external medium and those diffusing outward from the internal medium. (Supported by predoctoral fellowships awarded by the U.S.P.H.S. (HF 11, 368), and by a grant (G-12472) from the N.S.F.)

52

JOHN MAXWELL ANDERSON, Cornell University. Structural peculiarities of the pyloric caeca in a particle-feeding sea-star, *Porania pulvillus*. (10 min.)

The highly specialized digestive system of the sea-star *Henricia* has recently been described in detail, with particular attention to the elaborate flagellary pumping organs (Tiedemann's pouches) associated with the pyloric caeca, and the probable relationship of these specializations to flagellary-mucous particle-feeding pointed out (Anderson, 1960: Biol. Bull., 119). A similarly detailed examination of the digestive system in *Porania* is of interest in view of Gemmill's demonstration (1915: Proc. Zool. Soc. London) that this sea-star can maintain itself exclusively by particle feeding; we apparently have no information to indicate whether internal adaptations to particle-feeding in *Porania* are like those characteristic of *Henricia*. Preliminary anatomical and histological studies have been made on specimens generously provided by the Biological Station, Espegrend, Norway. A deep pouch occupies the oral face of each pyloric caecum; its side walls, most interestingly, give off tapering secondary pouches, alternately to right and left, which

hang from the median ducts of the aboral glandular branches. Thus, in effect, each of the glandular branches given off from the main median duct of the caecum has its own small Tiedemann's pouch, correspondingly branching from the main median Tiedemann's pouch. The small pouches show vertical striations in their walls, formed by alternating stripes of "normal" tall, flagellated epithelial cells and crowded mucous gland cells. Corresponding stripes face each other across the narrow lumen, so that each small pouch provides a series of parallel, vertical flagellated channels leading upward into the glandular portion, separated from each other by bands of secretory cells. The continuous adhesions across the lumen between opposite secretory bands, so characteristic of Tiedemann's pouches in *Henricia*, are lacking in *Porania*. Branching of Tiedemann's pouches in *Porania* is a unique feature undoubtedly contributing to the efficiency with which ingested particles are distributed to the digestive chambers of the pyloric caeca. (Supported by Grant G-6007 from N.S.F.)

170

WARREN ANDREW, School of Medicine, Indiana University. A study of living blood of some species of Bermuda tunicates, as observed by phase microscopy. (30 min.)

Specimens of blood from tunicates have been observed over periods as long as 24 hours and the structure and behavior of the cells studied with the phase microscope. The species investigated have been *Ascidia nigra*, *Clavelina picta*, and *Ecteinascidia turbinata*. Nine or 10 types of blood cells are distinguishable in each of these species. The classification of these cells by earlier authors has been inconsistent in many instances and contradictory to some degree. The cells containing vanadium chromogens require little revision but some of the other forms, particularly the Q (quiescent) cells of Fulton, are renamed and redescribed, since the phase microscope reveals the presence of motile, beaded appendages on such cells. A classification of tunicate blood cells more in accord with that of blood cells and wandering tissue cells of the vertebrates is suggested. Many of the types of blood cells have their counterparts in fixed tissue cells in various parts of the body of these animals.

196

WALTER AUCLAIR, George Washington University School of Medicine. Monolayer cultures of *Rana pipiens* kidney cells.

A procedure has been developed for cell-sheet cultures from frog kidneys. These cultures have displayed vigorous growth and numerous mitoses over a period of three months.

Kidneys are dissected aseptically from pithed frogs, minced and washed in several rinses of calcium-free amphibian Ringer's (CFAR). The organs then are stored at 2-4°C for 16-24 hours in a 0.25-0.50% trypsin medium in CFAR containing glucose. After the period of refrigeration, the kidneys are stirred magnetically 1-3 hours in fresh 0.25-0.50% trypsin solution.

The resulting cell suspension is decanted into centrifuge tubes and spun 5 minutes at 1000 × G. The supernate is removed, a few drops of CFAR are added to each tube and the cell pellets are dispersed and pooled for a cell count. Approximately 10^6 cells per kidney are usually obtained. Culture dishes containing 12 mm cover slips (for later mounting onto Romicron Maximow slides or fixation) are seeded with the cell suspension and enough culture medium is added for a final dilution of 5×10^5 cells/ml.

The culture medium is: NCTC 109, 100 ml of which is diluted with 54 ml double-distilled water—10 ml; calf serum—2.0 ml; chick embryo extract—2.0 ml; 5% lactalbumin hydrolysate in amphibian Ringer's—1.0 ml; glutamine (100 mM)—0.2 ml; and penicillin, about 500 units per ml of medium.

Extensive cell sheets form within 2-4 days at 25° C and the cultures subsequently are maintained by replacing the medium (excluding the lactalbumin hydrolysate) every 3-4 days. (Supported by U.S.P.H.S. Post-doctoral Research Fellowship CF 12, 861.)

141

ROBERT AUERBACH, University of Wisconsin. Effects of transplantation on thymus lymphoid differentiation. (15 min.)

Thymus rudiments from 12-day embryos of C, C_3H and C × C_3H matings and from 13-day (12-day equivalent) embryos of AKR matings were implanted in the anterior eye chamber of adult male C, C_3H, C × C_3H, AKR, and C_3H × AKR mice to obtain a spectrum of donor-host combinations. Grafts were recovered after one week. In all situations involving the implantation of inbred-line thymus in its own strain (17/17) or in an F_1 hybrid of that strain (25/25), as well as of F_1 hybrid thymus in a similar F_1 hybrid host (100/100) typical lymphoid differentiation occurred. In all combinations in which thymus from one strain was implanted into an unrelated strain (26/30) or in which thymus from an F_1 hybrid was implanted into either parental strain (10/11) or an unrelated strain (37/41) lymphoid differentiation failed to ensue. Instead, grafts developed into disorganized, large masses comprised predominantly of epithelial and fibrous material. Since other embryonic tissues grow and differentiate readily under similar conditions, the results require interpretation. It is suggested that the explanation may lie in the loss of tissue adhesion concomitant with lymphoid differentiation, that this constitutes a release from tissue-level control mechanisms normally associated with differentiation and that this permits different control mechanisms to become operative or manifest. A hypothesis is developed which states that the thymus may represent the major primordium of the mammalian immunological system. (Supported by grant C-3985 from the National Cancer Institute.)

111

JOSEPH T. BAGNARA, University of Arizona. Onset of pineal and hypophyseal regulation of melanophores in *Xenopus*. (15 min.)

Recently it has been observed that the pineal apparatus is responsible for the body-lightening reaction which occurs in amphibian larvae when they are placed in the dark. On continuing these obser-

vations it was of interest to determine the onset of this reaction in *Xenopus laevis*. Melanization of melanoblasts commences at about stage 35 (stages of Nieuwkoop and Faber) and by this time these chromatophores are already hormone-sensitive. The few melanophores present on embryos of this stage contain dispersed melanin due to the action of the hypophysis. Melanophore contraction is evident if such embryos are decapitated or are deprived of their hypophyseal placodes. That the pineal is functional at stage 35 is indicated by the pronounced melanophore contraction which occurs when these embryos are placed in the dark for short periods of time. Furthermore, embryos and newly hatched larvae deprived of the pineal area of the brain fail to show melanophore contraction during similar periods in the dark. It is significant that melanophore contraction can be induced in these young embryos by immersing them in solutions of melatonin, the hormone presumably released by the pineal to evoke the body-lightening reaction. The results of these experiments indicate that melanophores of *Xenopus* are sensitive to both pineal and hypophyseal hormones at very early stages of development and lend further support to the concept that the pineal is indeed responsible for the body-lightening reaction of amphibian larvae. (Supported by grant G-6231 from N.S.F.)

197

DONALD BAIRD, Princeton University. A haplolepid fish fauna in the Early Pennsylvanian of Nova Scotia.

A skull roof of the palaeoniscoid fish *Haplolepis (Parahaplolepis)* aff. *H. anglica* from the upper Parrsboro formation (Riversdale group, Lower Pennsylvanian) near Parrsboro, Nova Scotia, provides the earliest record of the Haplolepidae and the the first from Canada. It shows close affinities with fish from the Low Main coal of Newsham, England. Time range of the subgenus (now extended) is from middle Westphalian A to late Westphalian B, *i.e.* most of the first half of Pennsylvanian time. Analysis of variation in dermal sculpture pattern within a population of *Haplolepis (P.) tuberculata* from Linton, Ohio, provides criteria for evaluating differences in sculpture in other haplolepids.

The associated fauna includes palaeoniscoids *(Elonichthys)*, crossopterygians *(Megalichthys, Strepsodus)* and fresh-water arthropods *(Anthrapalaemon, Palaeocaris)*. A small *Gyracanthus* spine confirms the survival of this acanthodian genus into Pennsylvanian time in North America. (Supported by grants from the N.S.F. and the W. B. Scott Research Fund, in cooperation with the Nova Scotia Museum of Science.)

75

CLINTON L. BAKER, Southwestern College. Spermatozoa of *Amphiuma tridactylum:* morphology, helical motility and reversibility. (Motion picture, 15 min.)

Large elongated spermatozoa (258μ) have slightly curved needle-shaped acrosome (10μ), head (67μ), neck (3.5μ) and tail with middle-principal piece (88μ), end piece (77μ) and tail piece (12μ). Axial filament extends from neck to tail piece; first as heavy rod soon flattening with dorsal enlarged band and ventral smaller band connected by thin, almost transparent bar (dumbbell in section) enclosed in cell membrane or sheath.

Flagellum arises from basal-body in base of neck, extends entire length of tail as planar coil on dorsal side, terminating in unsheathed tail piece. Flagellum is enclosed in cytoplasmic sheath which forms undulating membrane with flagellum as outer margin. Middle-principal piece contains numerous mitochondria of characteristic shape closely applied to thin bar of axial filament (electron microscope); reacts to tetrazolium chloride by forming formazan indicating succinic dehydrogenase and ATP as source of energy for motility.

Fixed sperms are typically planar flattened while motile ones are invariably helical and spiral clockwise in forward motion. Apparent planar lashing of body is actually three dimensional rotation with no apparent movement of any component other than the planar vibratile flagellum. Tip of acrosome and tail piece constitute axis with latter acting as rudder.

Motion pictures clearly illustrate this helical motion. Protoplasmic blobs attached to tail move clockwise. Sperms encountering obstacles show reverse flagellar beat, direction and rotation. Reverse motility seems efficient and directive for change of direction. (Supported by grant RG-7209 from the U.S.P.H.S.)

12

DAVID F. BALPH, Utah State University. The behavioral response of free Uinta ground squirrels *(Citellus armatus)* to a trap. (15 min.)

The objective of this study was to investigate the role of behavior in trapping and retrapping Uinta ground squirrels.

The research was based upon observing and recording activity of known ground squirrels near a trap. Records were kept on variables associated with their movements and behavior patterns within a 6-foot radius of a trap. We hoped that comparing data among and within individuals would shed light on learning and other variables in an animal's response to a trap.

The data indicate that once an animal is captured its behavior reflects the trap as a source of reward (bait) and punishment (capture). If the animal is not captured on the first few approaches to the trap, subsequent approaches are often modified by what occurred on previous encounters. (Supported by Utah State University, Division of University Research.)

65

EDWIN M. BANKS, A. V. NALBANDOV and W. H. JOHNSON, University of Illinois. Motor components of sexual behavior in sheep. (Motion picture, 15 min.)

The action patterns displayed by sheep in sexual encounters were studied under pasture and small pen conditions. Data were recorded by means of cinematography, polygraph and tape-recorder. Preliminary analysis of the male pattern revealed 9 components, 7 of which appeared to be appetitive

in nature: approach, smelling perineal region, nudging, smelling urine, stepping back, mounting and intromission. The consummatory complex consisted of pelvic oscillations and ejaculation. Post-consummatory activity other than dismounting was variable.

Female sexual behavior reflected cyclic ovarian changes. Anestrous ewes responded to male approach with indifference or active avoidance. During early estrus, low intensity drive was characterized by standing and turning the head back to watch the male; vigorous males were accepted. Later, high intensity responses included active soliciting by smelling the scrotal and perineal regions, and by a circling movement around the ram; these led to male acceptance. Prior to the termination of estrus, low intensity components reappeared and were followed by the indifference or avoidance responses of the anestrous ewe.

Detection of an estrous ewe by the ram is a function of visual and olfactory cues. When placed into a flock of ewes, only one of which was in estrus, experienced rams rendered anosmic electrolytically successfully detected and copulated with her; this discrimination required substantially more time than that used by normal males in the same situation. (Supported by grant M-3203 from the U.S.P.H.S.)

11

KYLE R. BARBEHENN, National Institute of Mental Health. Interspecific spatial relationships among small mammals. (15 min.)

Recent field studies have indicated that certain small mammals (*e.g. Sorex cincereus, Pitymys pinetorum*) show a pronounced lag in entering snap traps when removal trapping is continued for a long period of time (15-30 days). A possible cause of this behavior may be that some species (*e.g. Peromyscus leucopus*) are psychologically dominant to others and their presence inhibits the use of space (and exposure to traps) by the subordinate forms. In a field experiment, *Peromyscus* greatly reduced the utilization of a bait station by *Microtus pennsylvanicus*. The altered patterns of movement by the *Peromyscus* in response to the bait station disrupted the behavior of the *Sorex* such that they were caught in relatively large numbers from the beginning of removal trapping, in contrast to unbaited control areas. Laboratory tests are now being conducted to measure interspecific influences on the utilization of space and to determine the associated mechanisms. (Supported by fellowship MF-9528 from the National Institute of Mental Health, U.S.P.H.S.)

93

STEPHANIE H. BARCH, Michigan State University. Serological analysis of Lucké renal adenocarcinoma. (15 min.)

As part of a research project attempting to analyze the mode of transmission of the Lucké renal adenocarcinoma in the leopard frog, *Rana pipiens*, a series of serological tests has been performed, employing the Ouchterlony agar-diffusion technique, in which antisera developed in rabbits against tumor and normal kidney tissue were reacted with antigens prepared from various adult frog tissues. All antisera were obtained by means of the Freund adjuvant technique. Appropriate control sera were tested prior to each immunization. Tumor tissue varied somewhat in regard to the total mass of carcinomatous growth found in individual animals, and in the degree of replacement by the tumor of normal kidney. Results from testing 13 antisera against tumor, and 12 against normal kidney tissue are reported. There was some variation in the numbers of components visualized on agar-diffusion plates among the antisera prepared against the same antigen (tumor or kidney). In some cases, there were no distinguishable differences between anti-kidney and anti-tumor sera; in no case was it possible to remove all the kidney components from an anti-tumor serum and have left only 'specific' tumor components, by the usual absorptive techniques. It became apparent as more antisera were tested, however, that a consistent difference existed between the kidney and tumor tissues, namely, that the former has a component not found in the latter. The observation that some samples of tumor produced faint indications of this component and others did not show it at all, suggests that the varying content in tumors, of normal kidney cells may produce the variation in the precipitation lines. These results lead to a working hypothesis that, in the transformation of kidney cells to cells of the Lucké renal adenocarcinoma, certain antigenic components either disappear, or become incapable of eliciting an immune response. (Supported by grant C-3124 from the U.S.P.H.S. and American Cancer Society Institutional Research Grant to Michigan State University.)

61

GEORGE W. BARLOW, University of Illinois. Color patterns as indicators of motivation in the teleost fish *Badis badis*. (15 min.)

The important components of the color pattern in *badis* are the general body colors, body bars, eye-slash, and three ocelli. The color patterns can be discussed under 11 more or less typical manifestations. Each pattern occurs in one or more characteristic situations, some occurring under a wider range of circumstances than others.

Pronounced fright is correlated with the palest coloration, moderate fright with thin disruptive bars, and increasing aggressiveness with distinct markings such as broad bars and dark fins, and darker coloration. Sexually motivated males are black, females pale, colors associated with extreme aggressiveness, and fright, respectively. When actually spawning in the burrow held by the male, the female is dusky brown with the double eye-slash and the dorsal and caudal ocelli present. Manifesting the dorsal and caudal ocelli in other circumstances, usually seen against a pale background color, seems to indicate "without territory." Parental males are dark, but seldom black.

26

AUDREY BARNETT, Indiana University. The inheritance of mating type and cycling in *Paramecium multimicronucleatum*, syngen 2. (Introduced by F. Payne) (15 min.)

Most stocks of *Paramecium multimicronucleatum*, syngen 2, show a daily cycle or circadian rhythm of mating type reversals. The phase of the cycle (*i.e.* time after experimental "dawn" when the two reversals of mating type—III to IV and IV to III—occur) varies from stock to stock, but is inherited vegetatively under set conditions. Some stocks, however, do not cycle. Cycleless stock 30, which expresses mating type IV continuously, was crossed to cycling stock 11 when it expressed mating type III. Only one of 120 F1 clones survived: it cycled but had a phase differing from that of parental stock 11. This clone was inbred to yield 49 viable F2 clones. Of these 35 cycled and 14 did not, suggesting a 3:1 ratio and a single gene locus for control of the cycle. Genic control is also indicated by the agreement between coconjugant F2 clones. Of the 17 pairs in which both members produced exconjugant clones, 11 yielded 2 cycling clones and 6 yielded 2 cycleless clones. The cyclers showed a variety of phases and among the cycleless some were type III and some were type IV. These variations appear to be caryonidally inherited, *i.e.* they are determined by macronuclear differentiations. Evidence for this is derived from the detailed study of 48 expanded subcaryonides from 4 cycling and 2 cycleless clones. With two possible exceptions, there were no differences between the lines of the expanded subcaryonides, but independently expanded subcaryonides of the same clone did show differences. (Supported by a U.S.P.H.S. predoctoral fellowship of the Division of General Medical Sciences and by grant AT(11-1)-235 to T. M. Sonneborn from the A.E.C.)

29

JAMES M. BARRETT and SISTER DOMINIC MARIE OLINGER, Marquette University. An immunochemical test of the reliability of nuclear morphology as a taxonomic criterion for actinophryid Protozoa. (15 min.)

Although nuclear morphology is ordinarily regarded as an important basis for separating protozoan genera, it is sometimes ignored in favor of cell-body structure. It seems useful, therefore, to compare selected genera immunochemically with a view to testing the relative reliability of these two traditional criteria by this somewhat more objective method.

Three species of Heliozoa were compared: *Actinosphaerium nucleofilum, Actinosphaerium eichhorni* and *Actinophrys sol*. They were chosen because, although the two actinosphaeria clearly belong to the same genus on the basis of their cell-body morphology and multinucleate condition, the nuclei of *Actinosphaerium nucleofilum* differ markedly from those of *A. eichhorni*, while being almost identical in appearance to the single nucleus of the smaller *Actinophrys sol*. The heliozoans were all cultured in 0.01% Knop's solution at 20-25° C and fed *Polytoma uvella* and a live bacterium found to support good growth of all three species. Male New Hampshire Red chickens served as antibody producers. Antisera were prepared against *A. sol* and *A. nucleofilum* by intravenous injections of lyophylized lipid-free cell homogenates resuspended in 1% saline. The antisera were tested with all three heliozoans by the modified micro double-diffusion technique of Crowle ('58), using appropriate absorption procedures.

The study showed that *A. nucleofilum* and *A. sol* share more diffusible antigens with each other than either does with *A. eichhorni*. On the basis that such antigenic similarity is reflective of kinship, *A. nucleofilum* and *A. sol* are more closely related to each other than either is to *A. eichhorni*. Thus, at least in the present instance, antigenic similarity parallels similarity of nuclear morphology and not of cell-body structure. (Supported by grants C-3890 (C1 & C2) from the U.S.P.H.S. and the Marquette University Committee on Research.)

74

H. W. BEAMS and EVERETT ANDERSON, State University of Iowa. Fine structure of "intracellular ductules" in certain glands of the carabid beetle. (10 min.)

It has long been known that certain insect gland cells, especially those producing an odoriferous or poisonous secretion, possess so-called intracellular ductules. One such striking example is found in the odoriferous glands of the carabid beetle (*Carabus* sp.) which are of the acinar type and composed of long (100 μ), narrow (25 μ), radially disposed cells. Each cell possesses an invaginated surface layer giving rise to a ductule which extends from the apex basally along the longitudinal axis of the cell, and as it approaches the base it winds around the nucleus, ending blindly in this region.

The ultramicroscopic anatomy of the ductule reveals the following structure arranged from the apical to the basal end of the cell as follows: (1) a short (18 to 20 μ) region composed of both epidermal and cuticle layers; (2) a small microvilli-lined enlargement or reservoir from which extends (3) a long ductule, the surface of which is studded with a multitude of microvilli; these bear a close resemblance to the surface (striated border) of certain other types of cells (*e.g.*, kidney; intestine). The ductule appears to be an adaptation on the part of the cell to greatly increase its polarized active surface area. If true, the term intracellular ductule is a misnomer as it is never within the cytoplasm, but always separated from it by the plasma membrane. However, the term is retained here, for want of a better one, to describe the interesting differentiation of this cell. (Supported by grants RG-4706 and 5479 from the U.S.P.H.S., and grant G-9879 from the N.S.F.)

48

R. M. BENOLKEN, University of Minnesota. Recovery processes in the eye. (Introduced by N. T. Spratt, Jr.) (15 min.)

Experimental evidence has been consistent with the hypothesis that the graded receptor potential, recorded intracellularly in the excised eye of *Limulus*, is the primary generator of impulse activity in the optic nerve. A model of the receptor membrane has been proposed on the hypothesis that the receptor potential arises as the result of light reactions which produce specific ionic permeability changes in the receptor membrane. Since a net

passive transport of ionic charge across the membrane would occur during the light response, this hypothesis indirectly requires recovery processes, such as ion pumps, which could restore the concentration gradients across the membrane after the eye has been stimulated by light.

Characteristic changes in the so-called resting membrane potential have been recorded in single photoreceptor units in the recovery periods which follow stimulation of the *Limulus* eye. It is suggested that these changes in the resting potential are electrical signs of predicted recovery processes. The data which support this suggestion will be presented and these data will be discussed in terms of the permeability model of the photoreceptor membrane. (Supported by grant B-2660 from the U.S.P.H.S.)

166

G. R. BERNARD, Medical College of Georgia. Some alterations of the adrenal glands of *Mesocricetus auratus* infected with *Trichinella spiralis*. (12 min.)

The adrenal glands of 172 infected (dose: 225 larvae) and 47 control golden hamsters were studied. A group of infected animals was killed and adrenals taken each week for the first 10 weeks and 3, 4, 6, 8 and 10 months post infection (PI). Adrenals from control animals were obtained at monthly intervals. All the hamsters were 8-10 weeks old at the beginning of the experiment and were housed in individual cages. NORMAL: The normal male hamster's adrenals were significantly heavier, showed significantly less ascorbic acid and non-specific dehydrogenase activity, with slightly more (but still very little) sudanophilic substance (s), and a much greater alkaline glycerophosphatase activity than the normal female adrenals. With increasing age there was a noticeable increase both in the cortical sudanophilic lipids and birefringence and a slight increase in ascorbic acid. Relative adrenal weights (mg/g body weight) did not change with age, but female adrenals were heaviest at 5 months. INFECTED: In 2 of 5 cases, 1 and 2 weeks PI, the alkaline phosphatase reaction was slightly more intense. Dehydrogenase activity was significantly raised during weeks 2 and 3 PI. Adrenals were heavier during weeks 3-9 PI. Ascorbic acid was significantly elevated 6 weeks PI. In both sexes its concentration in the infected animals' adrenals was higher than normal. The concentration of sudanophilic droplets was slightly greater in the more severely affected cases than in control or less severely affected hamsters, but birefringence activity remained unchanged.

198

G. R. BERNARD and J. M. WINNER, Medical College of Georgia and University of Notre Dame. Some alterations of adrenal glands of *Mesocricetus auratus* during the course of infection with the Khartoum strain of *Leishmania donovani*.

In our experience intraperitoneal injection of homogenates of infected hamster spleens resulted in death of the hamster host during the third month post infection (PI). Group I consisted of 8-10 week old male hamsters; Group II, of male animals about a year old. Experimental and control animals from both groups were sacrificed on days 12, 23, 34 and 48 PI. Adrenals were weighed individually; the left was fixed for lipid studies and the right was assayed for its ascorbic acid content. Adrenal weights of infected animals began to increase between days 12 and 23 PI, but the increase was not linearly progressive. In all experimental animals, adrenal ascorbic acid content was depleted absolutely and relatively. Range of values was 0.41-1.43 mg/g adrenal tissue (6.3-11.4γ) for infected animals *vs.* 1.06-2.78 mg/g adrenal tissue (11.6-18.0γ) for control glands. In 74% of all glands studied small sudanophilic droplets were found in the glomerulosa. The remainder of the cortex usually took a background "wash" with greater depth of color centrally. About 80% of infected animals of Group I showed moderate lipid granule accumulations in the other cortical layers; 20% of the controls showed similar accumulations. Group II results were inconclusive although accumulations of lipid droplets were more striking in infected animals. While in many individual adrenals birefringence and sudanophilia were quantitatively similar, no conclusions could be drawn from the data.

70

WESLEY J. BIRGE, University of Illinois. Cytoplasmic RNA in the parenchymal liver cells of the hibernating ground squirrel *(Citellus tridecemlineatus)*. (15 min.)

The principal point of concern in this investigation rests with gaining some measure of comparative levels of ribonucleic acid (RNA) metabolism and protein synthetic activity in the liver parenchymal cells of active and hibernating ground squirrels. The initial approach to this problem, based on the microspectrophotometric quantification of the cytoplasmic RNA in the liver tissue of active and hibernating forms, is summarized herein. Two-wavelength absorption measurements were made on toluidin blue-nucleic acid preparations. Tentative results clearly indicate a decided drop in the cytoplasmic RNA content of liver cells commensurate with hibernation induced by cold treatment. With complete inactivation of voluntary responses, cytoplasmic RNA consistently drops to a level approaching one-half or less of the quantity noted in normal, active squirrels. With the resumption of post-hibernation feeding, there is limited evidence to the effect that cytoplasmic RNA content rapidly approaches levels indicative of the active state. This suggests that with the resumption of activity, the recovery rate leading to a normal level of RNA metabolism is quite rapid.

Subject to final confirmation, such data presumably may serve as a *general* index to the comparative levels of RNA metabolism and protein synthetic activity in the liver tissue of squirrels in active and hibernating states. It is further suggested that such information may be of possible value in establishing a reference base useful in estimating comparative levels of metabolic activity during hibernation in different mammalian forms.

199

WESLEY J. BIRGE, University of Illinois. A cytochemical study of the differentiating ependyma in the chick embryo.

In this study consideration was given to the differentiation of the ependymal lining of the midbrain and diencephalon in chick embryos of 8 to 20 days. Primary concern was devoted to fluctuations in cellular ribonucleic acid (RNA) content occurring concomitantly with ependymal differentiation. Cellular RNA content was demonstrated by basic dye-binding methods.

During development much of the ependymal RNA is associated with cells involved in the active differentiation of ependymal processes. At the onset of this phase of differentiation, increases occur successively in nucleolar, nuclear and cytoplasmic RNA. Peak RNA content parallels the most active period of process differentiation. Presumably, the RNA activity thus evidenced is correlated with protein synthesis associated with process formation. As process differentiation nears the end point, cellular RNA content diminishes to a low level. Seemingly, well differentiated ependymal cells exhibit little capacity with respect to RNA metabolism and protein synthetic activity.

Throughout the roof area of much of the diencephalon and midbrain, the cells of the ependymal lining display a more simplified pattern of differentiation; process formation is much reduced and primitive epithelial features are evident through day 20. Throughout development these cells retain a high content of cytoplasmic RNA which apparently does not reflect activity associated with cell division or process formation. It is tentatively proposed that the RNA activity seemingly inherent in these cells is associated with the synthesis of a proteinaceous secretory product, reflecting a special functional role for the ependymal lining of this area.

97

HELEN BLAUVELT, LEONARD HERSHER and A. ULRIC MOORE, New York State College of Medicine and Cornell University. The social behavior of goats deprived of maternal and herd care. (15 min.)

This pilot study was designed to suggest the effect of maternal and herd care on the social development of the young. An experimental group (5) of kids was reared with social stimulation but no contact with adults after the third day postpartum. They were fed from a common source. Five matched controls lived in a typical farm herd environment each feeding from its mother. Differences between the groups in sociality, capacity to avoid unknown cues and nocive stimuli were predicted. The two groups were tested for response to a strange object, social contact and aggressive play, "waning of the response" to an "alarm cue," and motility during the diurnal cycle. Different patterns of social organization develop in the two groups. (Supported by a Ford Foundation Grant.)

200

JOSEPH B. BOATMAN, PATRICIA A. LASKEY and MARVIN J. RABINOVITZ, William H. Singer Memorial Research Laboratory of the Allegheny General Hospital, Pittsburgh. Relation of time and mass of incubated thyroid slices to I^{131} and Qo_2 metabolism.

Qo_2 utilization has been described as a function of slice thickness (J. Cascarano and B. W. Zweifach, J. Biophys. & Biochem. Cytol. 6, 215, 1959). 0.45-0.95 mm thick bovine thyroid slices were incubated at 5 to 240 minutes in beakers containing 0.4 μc I^{131} in 4 ml of Krebs-Ringer phosphate buffer under 100% O_2 at 38°C in a Dubinoff shaking incubator. Warburg O_2 measurements on duplicate slices were made at 38°C under 100% O_2. Slice thickness was altered by insertion of aluminum foil in a Stadie-Riggs microtome. Dry and wet weight Qo_2 of slices and water content and I^{131} uptake of incubated slices were calculated.

The quantity of I^{131} uptake was linear with time, expressed as I^{131} counts per minute/mg dry weight. At 0.45 mm thickness, values progressed from 5" to 240" from 125 to 1740 c/m, 0.55 mm from 430 to 2920 c/m, 0.65 mm from 354 to 2410 c/m, 0.75 mm from 480 to 1110 c/m, 0.85 mm from 252 to 948 c/m and at 0.95 mm, 176 to 860 c/m. Values for Qo_2 based on wet weight ranged between 0.167 and 0.283 μl O_2/hr/mg while mouse diaphragm values measured 0.702. On dry weight basis, values fell between 1.43 and 2.23, mouse diaphragm at 3.12, with no significant correlation to slice thickness.

It was concluded that penetration and trapping of I^{131} by thyroid slices were optimal between 0.45 and 0.65 mm thickness. Lack of Qo_2 differences reflected a less sensitive measurement of diffusion and metabolic gradients than I^{131} utilization. (Supported by grant A-3543 from the U.S.P.H.S.)

82

H. E. BOND, Biology Division, Oak Ridge National Laboratory. A sex-associated protein in liver tissues of the rat and its response to castration and hormone treatment. (Introduced by Norman G. Anderson) (15 min.)

A protein component, designated as a sex-associated protein. normally present in the male rat liver and absent from that of the female has been described previously (H. E. Bond, Biochem. Biophys. Res. Comm. 3: 53, 1960). Further studies concerning the chemical and physical evaluation of the protein have been performed, and the results will be presented together with the results of experimental manipulation of the protein following castration and hormone treatment. The protein disappears slowly from the male following castration and rapidly following castration and treatment with estradiol. The component also disappears from the intact male treated with estradiol. The castrated male which receives testosterone treatment retains nearly normal amounts of the protein component, and the female treated with the same material develops up to about half the amount of protein found in the normal male controls. The amount of sex-associated protein in the young male is very much less than that found in the adult. Protein

changes in the animals were investigated by using DEAE column chromatography and ultraviolet analyses of the column effluents. The findings together with the time and dose relationship will be discussed.

169

RICHARD A. BOOLOOTIAN, University of California, Los Angeles. Coelomocytes of echinoderms. (30 min.)

The cellular elements from the body fluid of 28 different species of echinoderms were studied by phase contrast microscopy. Fourteen types of corpuscular elements were identified and the distribution, properties, characteristics and, where possible, functions were determined.

Several types of coelomocytes were observed in the species examined here which had not been previously described, *e.g.* the red corpuscles of the sand dollar and crinoid, and the lobular corpuscles of the crinoid. Some of the coelomocytes formerly described were also found in the above species. Among these include the controversial bladder amebocytes in which the presence of a bladder has been questioned. In addition, a new cell type, herein called the reniform cell (kidney shape) has been found in a tropical echinoid species.

A greater diversity of cell types was observed in the coelomic fluid of the regular echinoids than in the irregular echinoids, asteroids, ophiuroids and crinoids.

54

BOVBJERG, RICHARD V., University of Iowa. Mechanisms isolating two species of crayfish. (15 min.)

The crayfish *Orconectes virilis* Hagen and *O. immunis* Hagen overlap extensively in range but rarely in habitat; the former occupy lake shores and streams and the latter ponds. This isolation was corroborated by field observations in northwest Iowa but habitat overlap was noted in sluggish streams with mud bottoms.

Experimental data indicate behavioral differences are important aspects of the isolation mechanisms. Response to substratum variables was studied as was the inter-species aggressive behavior. In large inter-species groups, *O. virilis* excluded *O. immunis* from the most suitable areas of the experimental habitat and in small groups was dominant in individual tension contacts. These data indicate possible biotic exclusion of the pond form, *O. immunis*, from the stream by the stream species, *O. virilis*.

36

EUGENE C. BOVEE and DAVID E. WILSON, University of Florida. An ameboid stage in the feeding behavior of the helioflagellate protozoan. *Dimorpha floridanis*. (15 min.)

The helioflagellate, *Dimorpha floridanis*, feeds in the heliozoan stage as the heliozoa ordinarily do, by engulfing lobopodially the flagellated bacteria and flagellated protozoa which have been trapped by sticky protoplasmic "beads" on the extended axopods. In the presence of massed flagellates in culture media, however, it retracts the axopods, and the globular body moves among the flagellates by ameboid movements, engulfing flagellates singly and sequentially. Whereas addition of fresh distilled water causes the heliozoan stage to convert to flagellate form, such treatment of the ameboid stage causes neither return to the heliozoan form nor conversion to the flagellate stage. Ameboid locomotion of the ameboid stages is accelerated, temporarily, by the addition of distilled water to the media containing such stages of *Dimorpha floridanis*. So far as the authors are aware neither transformations to the ameboid form nor assumption of ameboid movement are known for other species of helioflagellates, nor for heliozoa.

40

JAMES C. BRADDOCK, Z. I. BRADDOCK and H. RICHTER, Michigan State University. Form discrimination in the Siamese Fighting Fish. *Betta splendens*. (15 min.)

Male Siamese Fighting Fish will build bubble-nests under pieces of waxed paper floating on the surface of the water. The amount of nesting activity can then be measured in square centimeters. Circular, square, triangular and oblong forms having the same areas were presented in pairs. The fish exhibited a marked preference for the circular, oval, and triangular forms as opposed to the oblong. (Supported by the N.S.F. Undergraduate Research Program.)

201

OSMOND P. BRELAND and JOHN G. RIEMANN, University of Texas. Preliminary studies of spermatogenesis in the mosquito, *Culiseta inornata* Williston.

The studies have been made using the squash technique and aceto-orcein. So far as known, this is the first report on spermatogenesis in *C. inornata*. Most of the meiotic activities in this species occur near the end of the last larval stage. In certain other mosquitoes that have been studied, these activities apparently occur primarily in the pupal stage. The chromosome complement of *C. inornata* is similar to those of other culicine mosquitoes that have been studied; the 2n number is 6, with two large pairs and one somewhat smaller pair. There are no recognizable sex chromosomes.

Several characteristics of spermatogenesis in *C. inornata* are suggested by preliminary observations. These include: the absence of observable single chromosome strands in prophase I; the presence of chiasmata in metaphase I; the occurrence of an interphase between meiotic divisions I and II; polarization of the chromosome arms toward one side of the cell in prophase II; and the presence of a "comma" stage in which the tails of the young spermatids have a definite curl.

Detailed studies of spermatogenesis have been made upon very few species of mosquitoes, and workers differ in their descriptions of the process in the same species as well as among different species. Both similarities and differences occur in *C. inornata* compared with the descriptions of other species. So far as has been determined, neither the

polarization of the chromosome arms in prophase II, nor the occurrence of a "comma" stage has been reported in the spermatogenesis of any other species of mosquito. (Supported by grant E-3424 from the U.S.P.H.S., and by The University of Texas Research Institute.)

69

RUTH ALLEN BREMILLER, HISAKO O. YOKOYAMA and NAOMI LEMKEY, Medical School, Northwestern University. Phosphomonoesterases of the rat duodenum and heart. (15 min.)

The phosphomonoesterases were studied by histochemical and biochemical techniques. Homogenates were prepared with various media and optimal conditions were established for extraction of the enzymes and their storage. Quantitative determinations were made on the ability of these enzymes to split inorganic phosphate from the same substrates used for microscopic studies—sodium beta glycerophosphate and naphtholic esters. Activity was observed over a pH range of 4.5-10.0 using an ammediol-acetate buffer system for this range. The effects of the activators, Mn^{++} and Mg^{++}, and the inhibitors, formaldehyde and sodium fluoride, were observed at different pH levels.

Observations on microscopic sections were compared with those on zymograms (R. L. Hunter and C. L. Markert, Science 125:1294, 1957) obtained by means of starch gel electrophoresis. The influence of activators and inhibitors was observed with the naphtholic substrates at pH 5.5 and 8.6 both in the microscopic sections and the electrophoretically separated bands of phosphatases in the starch gels. These results were compared with quantitative data from homogenates. (Supported by Grant G-59-24 from the Life Insurance Medical Research Fund.)

159

W. R. BRENEMAN and FRANK J. ZELLER, Indiana University. P^{32} uptake by chick testes as an end-point for gonadotropin assay. (15 min.)

P^{32} uptake by testes of day-old chicks provides a sensitive end-point for gonadotropin assay. Gonadotropins were administered 24 and 12 hours before autopsy and P^{32} was injected 18 hours before autopsy. P^{32} uptake was calculated as counts per minute per mg of testis. Acetone-dried anterior pituitary glands were used but dosages were expressed as mg equivalents of fresh glands.

Significant increases were produced by total dosages of 0.15 mg equivalent of anterior-pituitary gland of 6 month-old cockerels and by 0.30 mg equivalent of gland of 3 month-old cockerels. Anterior pituitaries of pullets were less potent. Ovine LH and FSH preparations were very effective. LH produced significant increases at a 1.0 microgram dosage level and FSH gave significant increases at a 15.0 microgram dosage.

Pituitary glands of pullets and cockerels which received three days of treatment with estradiol and testosterone propionate produced a much greater increase in P^{32} uptake than did equivalent amounts of control pituitary glands. A plant extract, lithosperm, which has inhibitory action on gonadotropins *in vivo* and *in vitro*, was given to chicks for short periods of time. Anterior pituitary glands of lithosperm-treated chicks also produced a greater P^{32} uptake in testes of assay animals than did control glands. Results are interpreted as evidence for inhibition of gonadotropin release from the anterior pituitary by steroid hormones and by lithosperm. (Supported by grant C2415-C5 from U.S.P.H.S. Acknowledgement is made to the Endocrinology Study Section of N.I.H. for ovine gonadotropin preparations.)

175

ROBERT BRIGGS, JACQUES SIGNORET and R. R. HUMPHREY, Indiana University. Transplantation of genetically marked nuclei in the axolotl. (15 min.)

There are currently available in the axolotl several genes affecting development at times ranging from cleavage to larval stages. These genes would be extremely useful as markers in nuclear transplantation studies of nuclear differentiation. It is also possible that by the use of such genes the analysis of nuclear differentiation might eventually be refined to detect changes in the genes themselves, if they occur. In the past the exploitation of axolotls for work of this type has not been feasible because axolotl eggs, like those of other Urodeles, have given very poor results in nuclear transplantation experiments. The present paper presents an analysis of factors determining whether or not nuclear transfers succeed in this species. This analysis shows that successful transfers require 1) activation of the recipient egg by heat shock (35°C for 5 min.), 2) enucleation by ultraviolet irradiation, and 3) nuclear transfer at 2 to 4 hours after the heat shock. A method, based on this analysis, has been tested by transplanting blastula nuclei into activated and enucleated eggs. Ninety-six per cent of the eggs cleaved and 77% developed into complete blastulae. The majority of these blastulae (66%) developed to post-neurula stages. The validity of the method was tested by transplanting blastula nuclei carrying the dominant gene D (for black pigmentation pattern) into enucleated eggs from a recessive white (dd) female. A total of 37 larvae were obtained. All showed the black phenotype, proving that the nuclear transfers were successful. (Supported by grants RG-5850 from the U.S.P.H.S., and G-4284 from the N.S.F.)

64

BARBARA F. BROCKWAY, Cornell University. The effects of visual and vocal stimuli upon the reproductive ethology and physiology of paired Budgerigars *(Melopsittacus undulatus)*. (15 min.)

Females of this colonial species able to see and hear only their own mates rarely lay. Investigations were conducted to test the stimulatory effects of visual and vocal activities on reproductive physiology and behavior.

Pairs able to see and hear others (A), only hear others (B), only see others (C) and neither see nor hear others (D) were simultaneously used.

The relationship of the nest entrance to the perch is extremely important (entrances 2 inches below perches retarded nest box occupation and

egg-laying by at least 2 months compared to nest entrances 6 inches above perches). This had less effect on male reproductive behavior.

The amounts of male and female precopulatory behaviors were recorded for each individual per hour of observation throughout the experiment. The "warble" was the only male vocalization found to correlate with breeding. These behavioral activities are a more sensitive indicator of reproductive states than is gonadal inspection.

Initially, all males possessed sperm and all females had undeveloped ovocytes. Reproductive structures for all birds were preserved and weighed after one-quarter of the females had laid.

Results indicate that both sperm production and precopulatory activities were high in all groups but D. Group A scored highest with B and C about equal with two notable exceptions: B (hear only) "warbled" as much as A, and C (see only) females investigated and occupied nest boxes about as much as A. Only rarely, did low intensity male precopulatory behavior including "warble" occur in D. (Aided by grants from the American Museum's Frank M. Chapman Fund and the Sigma XI—RESA Research Fund.)

14

F. H. BRONSON and D. E. DAVIS, Pennsylvania State University. The effect of density and aggression on interaction rates in woodchuck populations. (15 min.)

Seven aggregates of five neighboring woodchucks in natural populations were marked and observed for three to four weeks each during 1959 and 1960. Data collected were: (1) aggressive interaction rate; (2) population density of each aggregate; and (3) proportion of activity consisting of various behaviors. Observations totaled 11,648 minutes of activity. Dominance testing in pens verified the relative social rank of each animal. Analyses of tolerance distances and interaction rates in the field showed that aggressiveness declined progressively after late April. Therefore, neighbor aggregates were observed at different levels of aggressive response as well as at different densities. Data were then analyzed to evaluate population density and behavioral indices as estimators of interaction rate. Density was not correlated with interaction rate unless combined with an aggressiveness factor. Behavioral indices correlated well with interaction rates.

104

VICTOR J. BROOKES, Oregon State University. Metabolism of the insect fat body. (15 min.)

The metabolism of variously labelled amino acids, acetate and glucose has been studied in intact and cell-free preparations of the fat body of the larvae of *Phormia regina*. Intact fat bodies were capable of extensive degradation of certain amino acids (valine, leucine and glutamic acid); as much as 60% of valine μ C^{14} and 85% of valine l C^{14} were recovered as $C^{14}O_2$. Little or no CO_2 production was found with arginine, histidine and methionine. Approximately 50% of acetate was converted to CO_2 but there was no CO_2 evolution when glucose was added to the incubation medium. Most of the activity with amino acids was lost when homogenates of tissues were used. What activity remained was found in the mitochondria. Amino acid metabolism was greatest during the growth of the larvae and dropped drastically early in the interval between the cessation of growth and the formation of the puparium. The fate of amino acid carbon will be discussed and some comparisons will be made between the metabolism in *Phormia* and the metabolism in other species of insects. (Supported by grant E-2390 (C2) from the U.S.P.H.S.)

202

IDELISA CALVENTI and ROSS F. NIGRELLI, New York Aquarium, New York Zoological Society. Effects of copper salts on *Fundulus heteroclitus*.

Copper acetate and copper citrate were non-toxic to male killifish at 0.4 ppm of copper for 18 days, although twice as much copper accumulated in the liver treated with copper citrate. Fish survived for three days in environmental concentration up to 1 ppm and for five days after single injections of 25 gammas of copper as copper sulfate or copper acetate. More than 50 gammas of copper caused death within 24 hours.

All treated fish showed some degree of congestion of the gills, liver and lower digestive tract. Increase in the respiratory rate at regular intervals, darkening of the skin accompanied usually by lateral flexion of the tail with absence of response to tactile stimuli, suggested a neurotoic effect of copper.

Fish exposed to higher concentrations of copper in water (1, 5, 10 ppm) or injected with 50, 75 and 100 gammas showed significant histological changes. Karyorrhexis and vacuolization of the liver cells were characteristic; kidney showed extreme granulocytopoietic activity and there was evidence of increased erythrocyte fragility. Further, eosinophilia persisted for about 72 hours when fish were immersed in solutions of 2 ppm, and for at least 24 hours after a single injection of 50 or 75 gammas of copper.

Of special interest in copper treated fish is the activation of latent bacterial infection with Gram-positive, non-motile, diphtheroid-like rods. The significance of this reaction is not fully understood.

203

GEORGE CAMOUGIS, LOUIS P. GRANATH and RICHARD G. BESCHLE, Clark University and Worcester Polytechnic Institute. Preliminary studies on the navigational ability of electric fishes.

In an exciting paper Lissmann (J. Exp. Biol. 35: 156, 1958) reported that certain electric fishes can discriminate between conductors and nonconductors placed in the surrounding water. To test whether this discrimination could act as a navigational aid in avoiding collisions with obstacles, we mounted two series of rods vertically in a small aquarium in which fish were free to swim. One series was made of wood, and the other of aluminum of the same thickness. Each series was supported on an input transducer which was activated by a collision with

the suspended rods. The transducers fed into a two-channel amplifying and counting system. The positions of the rods were interchanged systematically to cancel possible sensitivity differences or unknown differences due to position. No attempt was made to control ambient light intensity. Summations of collisions from several tests were used to compute collision ratios between metal and wood. These ratios were studied along with collision ratios for individual tests. The results indicate that *Hypopomus* collided with wood more often; *Gnathonemus* and *Gymnotus* collided with both equally often; *Sternopygus* tended to collide with metal more often. However, peculiarities in behavior may influence the collision ratios. For example, *Gymnotus* made repeated attempts to jump out of one particular end of the aquarium. These jumps caused most of the collisions. Several genera of nonelectric fish *(Pimelodella, Colisa* and *Anoptichthys)* collided equally often with both types of rods. Light gradients may influence collision ratios with all fishes.

20

GRACIELA C. CANDELAS, EVELINA ORTIZ, CARMEN VÁSQUEZ and LIVIA FÉLIX, University of Puerto Rico. Respiratory metabolism in tadpoles of *Leptodactylus albilabris*. (15 min.)

Field observations have revealed that tadpoles of *L. albilabris* will survive in moist clay for several weeks. In the laboratory they live for 40 days when kept in moist cotton. Tadpoles kept in this manner display lethargic behavior, and when these lethargic animals are restored to water they appear to regain their full activity immediately.

A total of 117 tadpoles in the premetamorphic stage were used, distributed about equally in water and in moist cotton. The animals were not fed during the experimental period, which lasted 3 to 40 days. Tadpoles in moist cotton lost weight faster and reached maximum weight-loss in about three weeks.

In trying to follow up to metabolic behavior of the animals in the lethargic state, O_2 consumption was measured in the Warburg apparatus. Measurements were made in air and water under a variety of conditions. Results indicate: 1) a depressed O_2 consumption in the lethargic tadpoles when kept in this condition in the flask; 2) lethargic tadpoles display a normal O_2 consumption within 45 minutes of exposure to water in the flask; 3) normal tadpoles fall into a depressed level of O_2 consumption within 45 minutes of withdrawal from water.

Further studies will be conducted to investigate the nature of the respiratory patterns in the lethargic stage in these tadpoles. (Supported by grant A-3628 from the U.S.P.H.S.)

63

BLAIR T. CARBAUGH and M. W. SCHEIN, Pennsylvania State University. Sexual response of roosters to full and partial models. (15 min.)

Twenty sexually experienced White Leghorn roosters that responded sexually to a taxidermic model of a receptive hen were used. Response levels were measured by assigning increasing values to each step in the sexual pattern: approach, grab, mount, tread, copulation. Percents of maximum possible levels during a 1-minute period were calculated and compared. A fully crouched model, used as a standard, elicited an average level of 78% of maximum; headless, tailless body, 56%; bodyless head with tail, 52% headless, bodyless tail, 23%; bodyless, tailless head, 43%. Eighteen males responded sexually to a crouched body without head or tail, 8 of the 18 were improperly oriented. In every case, where a head was present, orientations were correct. Thus, the head provides for orientation and has some arousal value, while the body has high arousal value but provides no orientation. These data will be compared to information on turkeys.

204

WALTER CHAVIN and MADELEINE OLIVEREAU, Wayne State University and Institut Océanographique. Adrenal histochemistry of fresh water and marine teleosts.

Recent histochemical findings have demonstrated that many substances characteristic of the higher vertebrate adrenal cortex were also present in the homologous tissue of goldfish. Extending these findings, this study deals with the histochemical patterns of the teleostean adrenal tissue in fresh water and marine species and correlation of such patterns with the activity of other endocrine glands.

Five to 24 animals of each of 7 fresh water and 5 marine species were utilized. Each fish was subjected to all test (lipid, phospholipid, cholesterol, ascorbic acid, chromaffin, norepinephrine, epinephrine-norepinephrine) and control measures. The animals were maintained for a minimum of 5 weeks and then rapidly sacrificed by decapitation when asleep or after electric immobilization, to avoid stress.

The adrenocortical (interrenal) tissues of all individuals of all species were positive for lipids, phospholipids, cholesterol and ascorbic acid. Marine species yielded more intense reactions for these substances than fresh water species. Individuals with the more active thyroid and indication of gonadal maturation also showed a more intense adrenocortical lipid reaction.

Despite similarities of adrenocortical cell size and structure in any one individual, the histochemical reaction was not uniform. Thus, morphological criteria are not always indicative of the physiological state of the cell.

Epinephrine-norepinephrine and norepinephrine were demonstrated in all species except *Anguilla anguilla,* in which the chromaffin reaction was not obtained even by the classic techniques. Because of the scattered nature of the chromaffin tissue, it was not possible to determine whether epinephrine and norepinephrine were present in separate cells. (This investigation was conducted under the auspices of a N.S.F. Senior Post-Doctoral Fellowship awarded to the senior author.)

126

JOHN R. COLEMAN, Carnegie Institution of Washington and Johns Hopkins University. Deoxyribonucleases in embryonic and adult *Rana*

pipiens. (Introduced by James D. Ebert) (15 min.)

Deoxyribonuclease activities (DNase I and II) have been followed in developing *Rana pipiens* embryos and in selected adult frog tissues. Enzyme activity was assayed by the spectrophotometric determination at 260 mμ of progressively increasing perchloric acid-soluble products resulting from degradation of DNA during incubation with homogenates at 27°C. Adult frog pancreas contains a nuclease of the DNase I category, requiring divalent cations and showing a pH optimum at 8.5-9.0, whereas liver exhibits activity of the DNase II type, inhibited by divalent ions and having a pH optimum at 4.5-5.0. Both types of activity are present in the tadpole, but undetectable in early developmental stages. Enzyme determinations on developing embryos at daily intervals show that detectable levels of DNase II activity first appear at or shortly before hatching. In contrast to DNase II, DNase I activity is low or absent until about the time of operculum closure, when it increases to a clearly detectable level. From the time of their appearance, both enzyme activities show pH and ionic requirements similar to those of the corresponding adult DNase I and DNase II. Possible regulatory mechanisms will be considered.

101

NICHOLAS E. COLLIAS, PETER J. FRUMKES, DONALD S. BROOKS and RONALD J. BARFIELD, University of California, Los Angeles. Nest-building and breeding behavior by castrated Village Weaverbirds *(Textor cucullatus).* (15 min.)

Castrate males develop a whitish bill as in non-breeding birds, but may attain high dominance, establish territories, sing, display and build nests. The black color of the bill, typical of breeding birds, was induced in a castrate by treatment with testosterone propionate. Daily injection of testosterone propionate into a non-breeding intact male and of gonadotrophins into three non-breeding intact males was accompanied by singing, pronounced rise in the dominance hierarchy, establishment of territory and increase in nest building activities. It appears that breeding behavior in this species depends to some degree on the gonads and is to some degree independent of the gonads. (Supported by N.S.F. grant G-9741. We wish to thank Thomas R. Howell for doing the operations.)

205

LOUIS D. DE GENNARO, Le Moyne College, Syracuse, N. Y. Chorioallantoic grafting of radioactive chick glycogen body.

White leghorn chick embryos received 5.0 μC glucose C-14 in 1.0 ml distilled water by injection into the air sac during incubation. The presence of radioactivity in the glycogen body, liver and heart was detected by autoradiography. Histochemical methods were used to stain and distinguish polysaccharides. Administration of radioactive glucose at 6 days resulted in strong concentrations of isotope in glycogen of the heart and in acid mucopolysaccharides of vertebral cartilage. Glucose C-14 was incorporated into glycogen of the glycogen body and liver when administered at 10 days of incubation. Radioactivity was present in the glycogen body and liver during the remainder of development and on the day of hatching.

Glycogen-labelled explants of liver, heart and synsacrum (with glycogen body) prepared from 12-day donors previously treated with glucose C-14 at 10 days were transplanted to chorioallantoic membranes of non-treated hosts. At recovery, some grafts of each tissue were void of radioactivity while some showed concentrations of isotope comparable to that found at the time of transplantation. Radioactivity was not found in well-vascularized grafts in which chorioallantoic blood vessels penetrated the transplanted tissue of the graft. Evidence to date indicates that radioactive glycogen is lost from chorioallantoic grafts of liver, heart and glycogen body by some influence of the vascular system and that radioactivity is found in the host. The results suggest that glycogen of the glycogen body, like that of the liver and heart, can be utilized by the chick embryo. (Supported by grant G-12902 from the N.S.F.)

91

LOUIS E. DeLANNEY, Wabash College. Homografting of sexually mature Mexican axolotls, *Siredon mexicanum.* (15 min.)

Recent reports of homograft rejection in mature and larval poikilotherms (fish, newts, frogs) have been described as comparable in mechanism to immunologically mediated rejections in homoiotherms. The possible existence of such capacity is particularly of interest in Mexican axolotls since the usual laboratory populations remain permanently aquatic although sexually mature and the question of immunological maturity can be raised. Most homografts between black and white strains, between the same strains, and in various combinations between sexes involved six sets of reciprocal grafts spaced one week apart. Graft rejections that have occurred to date show evidence of acceleration in later sets and the sequence of events, in order, involves excellent revascularization of the graft followed by hemostasis, hemorrhage, epidermal slough, and subsequent would repair. The criteria of rejection are more obviously demonstrated with black hosts bearing white grafts than the reciprocal graftings but these also may be rejected and at accelerated rate for second set and later implants. Some male animals of both black and white strains fail to reject grafts and the distinction between capacity or lack of capacity to reject is a quality of the host, not antigenicity of the graft, as a series of cross graftings demonstrates. Interpretations concerning the possible genetic, physiologic or endocrinologic aspects of these results, along with histologic studies of biopsies, will be discussed. (Supported by grant RG-5619 from the U.S.P.H.S.)

129

MARIE A. DiBERARDINO, Institute for Cancer Research, Philadelphia. The karyotype of *Rana pipiens* and investigation of its stability during embryonic differentiation. (Introduced by T. J. King) (15 min.)

In order to establish the karyotype of *Rana pipiens* and to investigate whether the karyotype changes during embryonic differentiation, metaphase chromosomes from the undifferentiated animal hemisphere cells of the early gastrula and from the differentiated cells of the post-neurula tail were studied in acetic orcein squash preparations. Thirty-three early gastrula diploid metaphases were analyzed, and were compared to eighteen post-neurula androgenetic haploid and four post-neurula diploid metaphases. The early gastrula analyses were derived from nine individuals arising from two sets of parents, and the post-neurula metaphases from twelve individuals arising from six sets of parents. In both cases the karyotype is the same and $2N = 26$. The chromosomes have been classified as follows. Group 1-4 with median centromeres are distinguishable by length differences; 1 and 2 are large; 3 and 4 are intermediate in size. Group 5-6, small chromosomes of similar length with median centromeres, can not be distinguished individually. Group 7-8 consist of large chromosomes with sub-median centromeres; 8 has a larger (long/short) arm ratio and sometimes has a constriction in the long arm (near the end distal to the centromere). Group 9-10 have sub-median centromeres; 9 is of intermediate length and 10 is a small chromosome with a prominent constriction in the long arm. Group 11-13 consist of small chromosomes of similar length with sub-median centromeres; 13 has the largest arm ratio; 11 and 12 are difficult to distinguish from one another. No evidence of a heteromorphic pair of (sex) chromosomes was detected. (Supported by grant No. 71 C from the A.C.S. awarded to Dr. T. J. King)

10

LEE R. DICE and PHILIP J. CLARK, University of Michigan, Michigan State University and Cranbrook Institute of Science. Variation in behavior and temperament among three races of peromyscus. (15 min.)

The behavioral responses of laboratory-bred prairie deermice, woodland deermice, and old-field mice *(Peromyscus maniculatus bairdi, P.m.gracilis,* and *P.p.polionotus)* were measured in a number of experimental situations. All the measures have a high degree of variability within each stock, as shown by coefficients of variation of 30 to more than 1000. The *bairdi* are relatively slow to enter strange situations, are likely to "freeze" for a time in a passageway, and have the highest coefficients of variation. The *polionotus* in comparison are bold and persistent and least variable of the three races. The *gracilis* are intermediate in boldness, persistence and variability, and exhibit a tendency to stop and explore the testing apparatus. (Supported by grants M-375, M-1650, and M-2050 from the U.S.P.H.S., and 923 from the Faculty Research Fund, University of Michigan.)

99

WILLIAM C. DILGER, Laboratory of Ornithology, Cornell University. Changes in nest-material carrying behavior of F_1 hybrids between *Agapornis roseicollis* and *A. personata fischeri* during three years. (15 min.)

The nest-material carrying of the same 6 female hybrids was studied from its onset to the end of the three-year period. Carrying in the bill *(fischeri)* and in the feathers of the back *(roseicollis)* are the two methods employed by the parental species.

The hybrids begin by spending much time attempting to carry by tucking, a method never successful for them. They manage to carry a small amount to the nest in their bills (6%). The next two months is marked by an improvement of the bill carrying method (to 41%). At this time the two methods are in strong conflict. The remainder of the time is associated with a slow but steady (about 10% a year) improvement in carrying in the bill and a coincident drop in behaviors associated with tucking. At the end of the period, over 73% are carried in the bill. Possible reasons are discussed. (Supported by grant G-14205 from the N.S.F.)

206

BURTON S. DORNFEST, JOSEPH LOBUE, EUGENE S. HANDLER, ALBERT S. GORDON and HENRY QUASTLER, New York University and Brookhaven National Laboratory. Further studies on leukocyte release from perfused isolated femurs of rats.

A technique for the assessment of bone marrow function in isolated rat femurs perfused with leukocyte-depleted blood has been described (Dornfest *et al.,* Anat. Rec., 137: 350, 1960). Direct evidence has been obtained indicating that the rate of leukocyte release from the marrow is influenced by the numbers of leukocytes in the perfusate and by the blood flow rate (Dornfest *et al.,* Fed. Proc., 20: 71, 1961). A positive correlation between flow rate and leukocyte discharge suggested that leukocytosis-inducing agents might operate by augmenting blood flow through the bone marrow. To test this, isolated femurs were perfused with blood containing the leukocytosis-inducing factor (Gordon *et al.,* Acta. Haemat., 23: 323, 1960). This resulted in marked increases in blood flow (5-8X) through the marrow and in the total numbers of leukocytes released (2-3X). Attempts to reproduce these effects with known vasoactive agents (acetylcholine, histamine, endotoxin) were unsuccessful. Perfusion of blood containing normal leukocyte numbers suppressed granulocyte release from the marrow. Mononuclear cells, however, appeared to have been sequestered within the marrow. Experiments to determine the fate of these mononuclear cells, the effects of varying the numbers of perfusing leukocytes and the influence of different perfusion media are now in progress. (A.S.G.—Supported by grant H-3357 from the U.S.P.H.S. B.S.D.—N.I.H. Postdoctoral Fellow. J.L. and E.S.H.—N.I.H. Predoctoral Fellows. Part of this work was performed at Brookhaven National Laboratory under the auspices of the U.S.A.E.C.)

207

ELLSWORTH C. DOUGHERTY, D. JoANNE FERRAL and BJORN SOLBERG*, University of

California, Berkeley. Xenic cultivation of antarctic micrometazoa. 1. The tardigrade *Hypsibius arcticus* (Murray, '07) Marcus, '28.

In April 1960, numerous anabiotic (desiccated) tardigrades *(H. arcticus)* were revived when algal "felt," collected in Dec. 1959 near Cape Crozier, Ross Island, Antarctica, was soaked overnight in distilled water. 6 individuals were isolated, 1/Bureau of Plant Industry watch glass (=B.P.I.—kept 6/Petri dish). Each aliquot of initial medium (see following abstract) received a fragment of 37-day-old blue-green algal film grown from "felt" in distilled water at *ca.* 18°C. The original cultures (also each set thereafter) were maintained, illuminated, at *ca.* 4°C except when examined at room temperature (usually *ca.* 17°C). 2 tardigrades yielded thriving xenic cultures, from 1 of which all subsequent cultures descend. In November the 2nd set was initiated with 6 animals (1/B.P.I.) in a modified medium: distilled water + xenic algal fragments from nitrate-free mineral medium; 6 excellent cultures resulted. 1 month later the 3rd set was started, with 5 eggs from 1 B.P.I. (same medium, but in 13 mm O.D. screw-capped tubes). All hatched; but the emergent nymphs, thereafter maturing, laying, and with 1 exception (to date) dying, have been variably fecund: 2, 14, 15, 53, 79 eggs (the last from the surviving parent, now again gravid), laid in clutches of 2-13 and isolated individually (as the 4th set—same medium). Hatching has been irregular: 0/2; 3/14; 9/15; 43/53; 47 (so far)/79; and subsequent survival to egg-laying, poor: 0/0; 0/3; 3/9; 1/43; 2 (so far)/47. Of the 6 egg-producers, 4 have proved fecund: 2 cultures are stored frozen, and 2 are being followed. (From 1 of the latter a 5th serial set has been started—in a medium similar to the earlier). Conditions giving improved hatchability and survival thereafter are now being sought. (Supported, in part, by N.S.F. grants G-9975 (U. S. Antarctic Research Program) and G-15034. *Present address: Shell Chemical Co., Pittsburg, California.)

208

ELLSWORTH C. DOUGHERTY, D. JoANNE FERRAL and BJORN SOLBERG*, University of California, Berkeley. Xenic cultivation of antarctic micrometazoa. 2. The rotifer *Philodina gregaria* Murray, '10.

In general, work with *P. gregaria* has closely paralleled that with *H. arcticus* (see preceding abstract), an important exception being that rotifers, brought back to California in the active state (by E.C.D.—see Dougherty, Chitwood, & Maggenti, Anat. Rec., 137: 350, 1960), have been subcultured serially since December 1959 (*i.e.*, without intervening anabiosis—however, others, of the same provenance as the tardigrades, were revived in April 1960, and lines descending therefrom are also current). Studies of fecundity, done with descendants of active animals collected in December 1959, have yielded, for the same media, results comparable to studies with tardigrades. The medium used for both organisms in April 1960 was based on algal fragments in aquarium water (a sample 2X sintered-glass-filtered, but unintentionally contaminated sometime before use; the aquarium itself had been started 1 year earlier, with water from a cow pasture stream, and had been periodically enriched with oven-dried rabbit-droppings). *P. gregaria* did well therein. In November, 5 immature rotifers (paralleling the 3rd tardigrade set) were isolated (1/tube) from 3 B.P.I. cultures set up in April. All 5 were relatively fertile, producing (viviparously): 12, 14, 20, 21, and 23 young; but these, isolated in turn, have shown considerably lower fecundity: reproducers/all progeny (so far)—4/12; 8/14; 10/20; 8/21; 16/23 (with a maximum number of 8 progeny—for only 2 animals).

A particular motivation for our working with *P. gregaria* (and *H. arcticus*) is the expectation that, properly studied, these organisms will reveal mechanisms of cytoplasmic inheritance. Certain of our data fit such an interpretation—but, at present, other explanations cannot be excluded. For a sufficiently profound approach, methods of synxenic and, ultimately, axenic cultivation are probably needed. (Supported, in part, by N.S.F. grants G-9975 (U. S. Antarctic Research Program) and G-15034. *Present address: Shell Chemical Co., Pittsburg, California.)

79

WM. G. DOWNS, JR. and DAVID FITZPATRICK, Tennessee Polytechnic Institute, Cookeville. Behavior of leucocytes and serum-proteins as a result of whole-body irradiation with large dosages of gamma rays. (15 min.)

Rats irradiated with dosages of gamma rays varying from 800 r to 1200 r show similar initial response in peripheral blood-count and bone marrow and leucocytogenic organs. There is an immediate marked fall in the lymphocytes and a rise in the granulocyte count, but a consistent fall in the total count, even in the first few hours. At the end of 24 hours all elements have decreased markedly.

In higher doses, *i.e.*, 1000 to 1200 r, the lymphocyte count falls to zero and the granulocyte falls as low as 100 per mm^3. From this period for several days no lymphocytes are seen in the circulating blood. They also disappear from the thymus, and frequently, in at least their normal appearance, from other lymphogenic organs. During this same period the beta and gamma globulin content rises somewhat, as measured by paper electrophoresis.

By the 12th to 14th day the total count rises, as do both the lymphocyte and especially the granulocyte count. By the 21st to 26th day the relative proportions of the two types of cells is nearly restored though the total count remains lower than normal for the animal. It requires about four months for the total and differential counts to return to approximately normal levels except in the rare cases when leukemia results. Blood protein fractions return to normal within a few days after irradiation.

103

CARBON C. DUBBS and RICHARD C. SANBORN, Purdue University. The osmotic pressure of the blood of individual insects. (15 min.)

Most determinations of the osmotic pressure of insect blood have been performed on pooled samples

from several animals. Since it is questionable whether or not insects are capable of regulating the osmotic pressure of their body fluids, we have measured the osmotic pressure of blood samples from single insects. We find that among animals from the same colony and with a common history of exposure to temperature and humidity, wide variations in the osmotic pressure of the body fluids appear.

As examples of these variations; adult *Periplaneta americana* reared at 28 ± 1° C and 62 ± 2% relative humidity have blood osmotic pressures ranging from 350 to 380 mosmoles/kg H_2O, diapausing *Samia cynthia* pupae reared together and stored in the same container during pupal life vary from 348 to 480 mosmoles/kg H_2O, and diapausing *Hyalophora cecropia*, also identically maintained, vary even more widely from 480 to 535 mosmoles/kg H_2O. Details of the variation in osmotic pressure when *Periplaneta americana* are reared at various relative humidities will be presented.

We are able to measure differences of less than 10 mosmoles/kg H_2O with samples of less than 10 μ liters of fluid by comparing the fugacity of blood samples with that of standard solutions when both are placed on thermistors in a Wheatstone bridge arrangement maintained at constant temperature. The method is sensitive enough to detect the polymerization of blood tyrosine into melanins upon exposure to oxygen. During the blackening of Cecropia blood, the osmotic pressure of the blood decreases by 50-70 mosmoles/kg H_2O within two minutes as a result of this process. (Aided by research grant G-11234 from the N.S.F.)

209

RICHARD M. EAKIN and JANE A. WESTFALL, University of California, Berkeley. Photoreceptors in the stirnorgan of tadpoles of the tree-frog, *Hyla regilla.*

Although there have been suggestions that the stirnorgan (frontal or Steida's organ) of amphibians is a light receptor no good evidence has heretofore been advanced in support of this hypothesis. Electron microscopy of the stirnorgan of young tadpoles of the Pacific Tree-frog, however, reveals processes extending into its tiny cavity which are similar to the outer segments of rods and cones of the vertebrate lateral eye and to those which we discovered in the third eye of the Western Fence Lizard. Each process in the stirnorgan of *Hyla* consists of a pile of 1-2 hundred or more discs or infoldings of the cell membrane along one side of the process, a feature characteristic of cones in the eye of *Xenopus* (Lanzavecchia, Arch. Ital. Anat. Embr., 65: 417, 1960). The process is joined to the cell proper by a connecting piece through which passes an apparatus of nine fibrils that terminates in an axial centriole. The distal end of the cell is packed with large ovoid or elongate mitochondria. These anatomical features strongly suggest that the stirnorgan is indeed a light receptor. Information received by it could be transmitted to the epiphysis along a nerve, which we have traced in *Hyla*. Studies are now being conducted to determine if the stirnorgan is involved in the control of melanophores (see Bagnara, Science, 132: 1481, 1960). (Supported by grant 14999 from the N.S.F. and by the Miller Institute for Research in the Basic Sciences of the University of California.)

55

ARLAN L. EDGAR, Alma College. Mating and oviposition in the phalangid *Leiobunum longipes* (Arachnoidea: Opiliones). (15 min.)

Mating in *Leiobunum longipes* may be divided conveniently into three phases: pre-copulatory, copulatory and post-copulatory activity. After the last molt, males and females assemble in "clusters" several feet above the ground on certain tree trunks. Attempts by males to copulate with females results in the gradual dispersal of these clusters. As time passes, males spend more and more time searching for females. When one is encountered in the presence of other males vigorous leg-pulling contests ensue. To copulate with a receptive female any male must successfully fend off the advances of other males in the vicinity. Curious tactile cues are used for rapid recognition. The female gradually becomes more receptive and, just prior to oviposition, will passively yield to any male of the same species. The male, by hovering near and often over the female, prevents her escape and "shepherds" her toward a stance of oviposition. Eggs are deposited in crevices of decaying wood, leaves and moist debris on the forest floor where they remain until they hatch the following spring.

The presence of females of other species causes no disturbance; males of larger species frequently attack *L. longipes* males and attempt to copulate with the females. In both cases, vigorous resistance is offered and escape is usually effected.

190

B. E. ELEFTHERIOU and M. X. ZARROW, Purdue University. The influence of thyroidectomy and thyroxine on brain maturation in two subspecies of *Peromyscus maniculatus.* (15 min.)

Determinations of total solids, water content, lipids, cholesterol and neurokeratin (trypsin-resistant) were carried out on the brains of two subspecies of *Peromyscus (P. m. gracilis* and *P. m. bairdii)* from the day of birth to 70 days of age. In the normally developing deermouse the brain weight of *P. m. gracilis* was significantly greater at all times than that of *P. m. bairdii;* however, with the exception of neurokeratin, the concentration of the above constituents was significantly greater in *P. m. bairdii.* Thyroidectomy, induced by injection of a total of 240 μc I^{131} over a 5-day period, caused a significant depression in brain weight and total solids, cholesterol and lipids, an increase in water content and no effect on neurokeratin. Both subspecies responded in a comparable manner. Treatment of the pregnant females and offspring with 1.2 μg thyroxine per kg of food led to an initial increase in brain weight and the various constituents, especially the lipids. By the 70th day of age, however, all values were within normal ranges. It may be concluded that thyroxine hastens and enhances maturation in both subspecies and that *P. m. bairdii,* which normally matures at a greater rate

than *P. m. gracilis*, is more sensitive to thyroid manipulation. (L-thyroxine was supplied by Dr. H. Fevold, Travenol Laboratories. Aided in part by a grant from the Purdue Research Foundation —2105.)

185

HANS ELIAS, Department of Anatomy, Chicago Medical School. Looking into space: a motion picture on three-dimensional interpretation of flat sections. (Motion picture, 40 min.)

Methods of reconstruction as well as statistico-geometrical methods for the determination of three-dimensional shape, size and volume distribution as applied in histology, paleontology, geology and astronomy are presented in a dynamic and instructive manner. (Supported by Grant RG-6533 from the U.S.P.H.S.)

28

ALFRED M. ELLIOTT, JOHN R. KENNEDY, JR. and IL JIN BAK, University of Michigan. Macronuclear events during mitosis in *Tetrahymena pyriformis*. (15 min.)

Synchronously dividing cultures of *T. pyriformis* (WH_6) were examined during 2 division cycles by light and electron microscopy. The cells were synchronized by subjecting them to heat shock treatment according to the method of Scherbaum and Zeuthen. Five ml samples were withdrawn from the flask before heat treatment was initiated and thereafter at 30 minute intervals for 7 hours during which time the cells went through two division cycles. The samples were fixed and prepared for both light and electron microscope examination. Treatment with appropriate enzymes and stains identified the DNA- and RNA-containing bodies during the different division stages.

Peripheral RNA bodies possess electron dense outer shells with less dense inner cores. During the second heat shock interval (approximately 1½ hours) centrally located RNA bodies appear which resemble the conventional mammalian nucleolus. They possess no membrane and are architecturally different from the peripheral RNA bodies. They reach their maximum size and density during the third heat shock after which they gradually disappear and are absent by the 4th heat shock (4 hours). Following this period the peripheral RNA bodies cluster near the membrane forming blebs which protrude into the cytoplasm. During the next hour these disappear and the cycle is initiated again. The DNA bodies double during the third heat shock and remain constant in number until cytokinesis occurs. Actual macronuclear and cytoplasmic division occurs about 60 minutes following the 5th heat shock. (Supported by grant E-1416 from the U.S.P.H.S.)

98

JOHN T. EMLEN, University of Wisconsin. Cliff edge responses of gull chicks. (15 min.)

Herring gull chicks tested on an elevated platform near their nest sites showed a clear avoidance of the edge and a strong reluctance to jump during the first three days post hatching. These negative responses declined with increasing age but at different rates in birds from different nest situations. Chicks from nest sites in precipitous cliff situations dropped the edge avoidance response more rapidly than chicks from plateau nests, but retained the reluctance to jump longer. Several possible explanations for the observed differences are discussed, and the proposal is advanced that different experiential backgrounds in the two nesting situations induce divergent developmental trends in the simple response to depth and the general characteristic of boldness towards unfamiliar situations. These characters are thought to combine to determine the cliff response of gull chicks.

210

LLEWELLYN T. EVANS, Research Laboratory, Jaffrey Center, New Hampshire. Aquatic courtship of the wood turtle, *Clemmys insculpta*.

A receptive female circles a male in water (9" or more in depth) with head turned, carapace tilted toward him. He lunges, gripping the edge of her carapace with his feet. Within seconds the floor of his mouth pulsates to pump water at the face of the female—averaging sometimes one pulsation per second. At intervals between pumpings the male extends his knees and elbows then quickly flexes them to bring his plastron and her carapace together with a loud clap. At irregular moments but following after the shell-clapping the male extends his head, open-jawed, then crashes it against her gulo-plastral plates to bite them in the downward stroke. This rhythmic water-pumping and staccato percussion might continue for three hours or even longer.

The male raises his head to the surface for air at approximate 15-minute intervals, whereas the female struggles violently to lift them both up for air every half hour or less. The male's movements also include the scraping of his shell upon hers laterally and postero-anteriorly. Finally his tempo rises to virtually a constant clapping and biting while the pumping stops. Soon the male flexes his knees, extends his elbows, thus sliding himself posteriorly to enable his tail to encircle hers thus achieving copulation. (The author is grateful to Mr. Norman Wight for his assistance.)

211

LLEWELLYN T. EVANS, Research Laboratory, Jaffrey Center, New Hampshire. An interpretation of turkey courtship.

Does the turkey follow the broad pattern of other Galliformes in adhering to a lek where the males gather and to which the females are attracted? Study at The Welder Wildlife Foundation* near Sinton, Texas enabled me to find a partial answer. The species *Meleagris gallopavo intermedia* Sennett inhabits Texas and adjoining territory. Mast of the live oak and other seeds and graze continue to attract the bird to this habitat. Three flocks occupied the Welder Refuge, each keeping closely to its circumscribed range of about 5 square miles, as indicated by banding records.

In February the flock (#1) of 40 birds becomes two monosexual groups of 20 each. The hens roost

in a separate area but near that of the gobblers. At pre-dawn the latter begin calling and fly down to feed. The hens appear to move parallel to the cocks but keep their distance, remaining hidden wherever possible. The males pass boldly seeking one of at least two leks which they occupy on alternate days. Here four large males chase and threaten the other 16 daily, causing them to leave the group, one by one to hide in the undergrowth. On April 1 these four definitive breeders begin a slow tour of the range, stopping momentarily to strut (usually by a single bird), then gobble in unison. The non-strutters, with heads high, gaze and listen (?). On April 4 they have separated and each calls *and* struts in different parts of the range. *M. g. i.* utilizes the lek to eliminate the weaker males. (*Kindness of Dr. Clarence Cottam, Director.)

154

R. M. EVANS, R. C. PURDIE and C. P. HICKMAN, Jr., University of Alberta. The effect of different temperature and photoperiod conditions on the respiratory metabolism of rainbow trout *(Salmo gairdneri).* (15 min.)

Mid-winter spawning rainbow trout *(Salmo gairdneri)* were acclimated for a minimum of 43 days to one of four temperature-light conditions: 16C-16L, 16C-8L, 8C-16L, 8C-8L. Oxygen consumption of the intact fish and of samples of liver, gill and brain were measured at acclimation temperature.

All tissues, except gill at 8C, showed a linear decrease in oxygen consumption with increased time of tissue storage. QO_2 values, corrected to a storage time of 15 minutes, showed a trend for the 8L tissues to metabolize more rapidly than the 16L group, except for gill at 16C. Photoperiod did not significantly affect total metabolic rates, but in the larger fish (40g) at 16C, the 8L group tended to show a higher metabolic rate than the 16L group.

Brain showed perfect metabolic compensation to temperature (QO_2 at $8° = QO_2$ at 16°) and liver showed overcompensation (QO_2 at $8° = QO_2$ at 16°). No compensation occurred in gill respiration, and total respiration showed partial temperature compensation. It is suggested that perfect compensation measured in brain would maintain nervous coordination and motor conduction at optimal levels, thus permitting a large degree of temperature independence of locomotor activity.

53

A. FARMANFARMAIAN, University of Shiraz, Iran and University of California, Berkeley. Storage and translocation of nutrients in the purple sea urchin *(Strongylocentrotus purpuratus).* (15 min.)

Recent investigations of the perivisceral fluid of the purple sea urchin indicated that certain organic constituents of this fluid are maintained essentially at a constant level throughout the year. The question arose, therefore, as to whether the level of nutrients in the perivisceral fluid is regulated by the animal, or the translocation of nutrients is achieved through the water vascular and hemal systems. The nature and site of stored nutrients and the mechanisms involved in nutrient translocation in echinoderms have been subject to much controversy and speculation. The present investigation was undertaken to clarify and settle these questions through the use of tracer techniques.

Sea urchins were fed $C^{14}O_2$-activated *Iridaea.* Conversion efficiency of active plant material into active animal material, and the distribution of activity in the tissues of the sea urchin were determined. These results indicate that the walls of the digestive tube, particularly of the festoon closest to the esophagus, should be regarded as the site of storage. In experiments where various parts of the water vascular and hemal systems were severed, it was shown that these systems do not contribute significantly to the transport of nutrients. Continuous sampling of the perivisceral fluid showed that the level of activity rises rapidly after a meal but subsides within a few hours to a low level where it is maintained. Active substances are found in both the plasma and the cells of the perivisceral fluid. It appears, therefore, that the level of nutrients in the perivisceral fluid is regulated. (Supported by N.S.F. grant G-10867.)

62

ROBERT W. FICKEN, Cornell University. Sexual behavior of the Common Grackle, *Quiscalus quiscula.* (Motion picture, 15 min.)

Sexual interactions between male and female Common Grackles were studied for three years under natural conditions. During the spring of 1961, stuffed females in the soliciting posture were placed near breeding colonies and the reactions of males to these were recorded. Marked differences in certain aspects of behavior occurred when males reacted to live versus stuffed females. Behavior elicited in the males by the stuffed females, but not by the live females, included attacks, light pecks especially at her eyes and cloacal area, lifting her bill, pulling her tail, preening and pulling out her feathers, unusual display forms, and successive mountings. Some suggested explanations of the differences in the reactions of the males will be given.

43

WARREN R. FLEMING, KENNETH SCHEFFEL and J. R. LINTON, University of Missouri. Cholinesterase activity in gills of several teleosts. (10 min.)

Cholinesterase activity in gill homogenates of several species of the genus *Fundulus,* and of the juvenile channel catfish, *Ictalurus punctatus punctatus* has been studied. The gill of *F. kansae,* a euryhaline teleost, contains the specific acetyl cholinesterase. The substrate optimum is 0.008 M, and activity is completely inhibited by eserine and the specific inhibitor BW 284C51. Eserine (10^{-6} M) inhibits sodium excretion by *F. kansae* held in hypertonic salt solutions for a short period of time and then the excretion rate returns to normal. Further addition of eserine invariably impairs opercular movement and the animals do not survive. Adaptation to a wide range of salinities does not change the activity of the enzyme significantly, but changes in the homogenates upon standing are observed, especially in fish which had been moved rapidly from tap water to 0.40 M NaCl. Gill homogenates

of *F. catenatus*, a stenohaline species, contain the non-specific cholinesterase in very small amounts. Homogenates from the catfish gill contain the specific enzyme with a substrate optimum at 0.004 M. The effect of cholinesterase inhibitors on salt excretion in this species is being studied. (Supported by N.S.F. Grant 13041.)

160

DARHL FOREMAN, Western Reserve University. Some short-term effects of gonadotrophins on the metabolism of the gonads. (15 min.)

Effects of gonadotrophins can be studied in two ways, by using either hypophysectomized animals or animals which have low endogenous titers of these hormones. This study has utilized both types, but has emphasized the latter by using immature rats and out-of-season prairie dogs. Both male and female prairie dogs are annual breeders with cyclic pituitary gonadotrophic activity.

FSH increased the oxygen uptake of ovaries from both prairie dogs and rats after injections into the animal and after *in vitro* additions of the hormone to Warburg flasks containing slices of tissue from untreated animals. This effect is probably due to effects on many enzyme systems, but does definitely involve the enzyme systems of the mitochondria. LH did not produce these effects when tested on ovaries which had not been previously treated with FSH.

Both FSH and LH decreased the ascorbic acid concentration of the right ovary in immature rats. This decrease occurred within one hour after the removal of the left ovary and injection of the hormone. These effects were not observed in hypophysectomized rats. FSH also produced an increase in the ascorbic acid concentration of immature rats 24 hours after the removal of the left ovary and injection of the hormone.

The oxygen uptake of the testis of the prairie dog is increased by small iv. doses of both FSH and LH. The doses given did not change testis weight. Prolactin increased oxygen uptake when added to the testis tissue in the Warburg flask. (Supported by grant A-3061 from the U.S.P.H.S.)

15

SISTER ALICE MARIE FOX, Clarke College. Active transport of sugar by turtle small intestine. (Introduced by X. J. Musacchia) (15 min.)

An *in vitro* preparation was used to demonstrate that ^{14}C-D-glucose is taken up at the mucosal surface of *Chrysemys picta* small intestine and moved to the serosal side. Uniformly labelled ^{14}C-D-glucose with a specific activity of 0.4 microcuries was the tracer.

Three types of experiments using the radioactive glucose were run: (1) ^{14}C-D-glucose was placed on both sides of the intestinal wall and incubated for one hour at 30°C (four segments); (2) ^{14}C-D-glucose was placed on the serosal side of the everted sac while the same concentration (2.5×10^{-4}M) of a non-radioactive glucose solution was placed on the mucosal side (four segments); (3) ^{14}C-D-glucose was placed on the mucosal side of the everted sac while non-radioactive glucose was placed on the serosal side. In this last way ten segments taken from active animals were run at 30°C, and ten segments taken from animals which had been in cold torpor were run at 2°C.

Movement of the sugar across the intestinal wall and its release into the serosal fluid against an apparent concentration difference was shown to take place at 30°C. The source of the sugar entering the medium on the serosal side appeared to be in part the glucose taken up from the mucosal fluid, and in part some stored carbohydrate. Tissue analysis using the anthrone reagent indicated the presence of a glycogen-like polysaccharide in the intestinal wall of both active and cold-torpid turtles. It was concluded that during intestinal transport in *Chrysemys picta* some of the glucose absorbed is converted to endogenous carbohydrate or metabolized, and some is translocated.

127

RONALD C. FRASER, University of Tennessee. The development of hemoglobins in the chick. (15 min.)

Following a preliminary analysis of the hemoglobins of 7- and 11-day chick embryos and the adult fowl by paper electrophoresis, a quantitative evaluation of the relative concentrations of the two types of hemoglobin in chick embryos of progressive age was undertaken.

There was found to be a complete reversal in dominant types of hemoglobin during ontogeny of the chicken. The embryonic hemoglobin found in concentration of 84% in the 5-day embryo was reduced during development to 28% by the time of hatching. This amount is retained in the adult.

The transition in hemoprotein concentrations did not parallel morphological changes in the red blood cell lines or sites of blood cell formation during the ontogeny of the chick. The progressive genetic expression of hemoglobin formation is considered to be carried through the changing hemopoietic foci during development. (Supported by grants E-171 from the American Cancer Society and G-3486 from the N.S.F.)

157

JOHN FREHN and ADAM ANTHONY, Pennsylvania State University. Respiration and phosphorylation of liver mitochondria from cold-exposed rats and chipmunks. (15 min.)

Comparisons were made of the effects of cold exposure (1 to 3 months at 1°C) on the respiration and phosphorylation of liver mitochondria in rats and chipmunks. Measurements were made polarographically using the oxygen electrode. The data indicate that the ADP:O ratio was slightly lower in mitochondria from cold-adapted rats using succinate and β-hydroxybutyrate as substrates; this ratio was unaltered in hibernating chipmunks. No differences were found in the ratio of the substrate-ADP respiratory rate to the substrate rate (R_s) in cold-exposed rats or in chipmunks that failed to hibernate. However, the R_s value of hibernating chipmunks was higher than that of controls. The adaptive significance of these changes is discussed.

212

W. L. FRENCH and J. B. KITZMILLER, University of Illinois. Cytological evidence for crossing-over in males of *Anopheles quadrimaculatus.*

Genetic crossovers have been reported in males of *Culex* mosquitoes (Gilchrist and Haldane, Laven) but no cytological substantiation is available. In *Anopheles* no genetic data are available due to the lack of mutants, but the present study reports cytological evidence of segmental interchange in *Anopheles quadrimaculatus.*

Salivary gland preparations from seven stocks of *A. quadrimaculatus* uniformly show a small paracentric inversion in chromosome III. Testis preparations stained with a modified aceto-orecin technique show anaphase I figures with dicentric chromosomes, bridges, looped chromosomes and acentric fragments indicative of crossing-over within an inversion.

In chromosome III the following configurations have been observed at anaphase I: (1) a single bridge with an acentric fragment probably due to a single crossover within the inversion; (2) a double bridge with two acentric fragments probably due to two crossovers within the inversion; (3) a looped chromosome probably due to a single crossover within the inversion and another crossover between the inversion and the centromere; and (4) two looped chromosomes and two fragments, probably derived from two crossovers within the inversion and another crossover between the inversion and the centromere.

Cross-shaped configurations in chromosome II (not translocations) are interpreted as resulting from single crossovers in one arm.

Testis chromosomes are in substantial agreement with salivary chromosomes with respect to relative lengths of the arms and position of the centromere. (Supported by grant E-3486 from the U.S.P.H.S.)

213

PAUL J. GARDNER and IRWIN L. BAIRD, Omaha University and University of Kansas. Some observations on venous flow in the turtle, *Pseudemys scripta.*

Routes traversed by blood returning to the heart from several sites have been studied radiographically in twelve specimens of *Pseudemys scripta.* Experimental animals were anesthetized by intraperitoneal injections of pentobarbital sodium. Continuous low-pressure injections of a contrast medium (Hypaque Sodium) were made into selected veins while radiographs were taken at intervals of five seconds each. Veins selected for injection included the femoral, ischiadic, caudal, external iliac, posterior vertebral, marginocostal and the first, third and fifth intercostal veins.

It has been suggested (Jourdain, 1859; Stromsten, 1905) that venous return from posterior somatic areas of the body occurs along two routes, one *via* a "renal portal system" and posterior vena cava, and the other *via* the ventral abdominal veins and hepatic sinusoids. Under the conditions of these experiments, only a small part of the flow from the posterior (dorsal) part of the external iliac vein, plus return from the posterior vertebral vein, enters the "renal portal system"; major return from posterior somatic structures is *via* the ventral abdominal veins. Conversely, excepting that of the first intercostal vein, the entire intercostal-vertebral drainage passes through a complex of small veins within the kidney and enters the posterior vena cava. Rapid injection into the intercostal-vertebral system results in reversal of flow in the renal portal vein and diversion of some of the contrast medium into the ventral abdominal veins; this tends to support speculation that direction of flow in the renal portal vein may vary with conditions of temperature and bodily activity.

162

RICHARD D. GILCHRIST and ROGER A. HOFFMAN, U. S. Army Chemical Research and Development Laboratories, Maryland. Changes in ascorbic acid and cholesterol in the cryptorchid rat testis. (Introduced by Ronald R. Novales) (15 min.)

Unilateral cryptorchidism in the rat is followed by a rapid and marked increase in concentration of cholesterol in the cryptorchid testis and little to no change in concentration in the scrotal testis. The concentration in the former reaches a maximum at 15 days and a return toward normal is evident at 30 days after operation. The total content of cholesterol in the cryptorchid testis, however, shows little change until 30 days. A significant reduction in the total cholesterol content of the cryptorchid testis is evident only at 30 days. When the amount of cholesterol in the cryptorchid testis is expressed as a percentage of the content in the scrotal testis, a straight line relationship is obtained in respect to time from 2 through 30 days.

Ascorbic acid is rapidly depleted following cryptorchidism. This occurs in the scrotal testis as well as in the cryptorchid testis, although to a much lesser degree. Concentration in the cryptorchid testis reaches a minimum at 15 days after operation and it then returns to normal by 30 days. Total amount per testis declines linearly with the testis to the minimum at 15 days with a partial return toward normal at 30 days.

No significant differences were found in total amount or concentration of ascorbic acid of the testis at 5 days between sham-operated and unilaterally or bilaterally cryptorchidized rats. Chronic ACTH or adrenalectomy has only a slight effect on these two components at 5 days.

17

ERWIN GOLDBERG, VIRGINIA W. PENCE and CHARLES NORMAN, West Virginia University. The distribution of free amino acids in cock and bull seminal plasma and spermatozoa. (15 min.)

The total concentration of free amino acids in bull seminal plasma is 172 mg per 100 ml and 406.9 mg per 100 ml in cock seminal plasma. This compares with a concentration of approximately 50 mg per 100 ml in the blood of most vertebrates.

Glutamic acid constitutes 43% of this total in the bull and 79% in the cock. The remainder of the pool in bull seminal plasma consists, in order of decreasing concentration, of arginine, aspartic acid,

alanine, serine, glycine, and trace amounts of phenylalanine, leucine and isoleucine. In cock plasma glumatic acid, glycine, arginine, alanine, and the others in trace amounts, are present.

In cock and bull spermatozoa differences in relative concentration of amino acids *vis-a-vis* cells and plasma, were observed. An approximation of cell volume suggests that most of the constituents of the free amino acid pool of the cells are in equilibrium with those of the plasma.

The physiologic significance of this high titer of amino acids is not yet known, although their contribution to the amino acid pool for protein synthesis is well-established in growing, dividing cells. Amino acids are, of course, potent buffers. The unusually high arginine concentration in cells and plasma may reflect the arginine-rich protein characteristic of sperm. However, it is interesting to speculate that this amino acid may be present as a phosphagen, providing energy for sperm motility. (Supported by grant RG-6339 (C-1) from the U.S.P.H.S.)

68

WILLIAM R. GOODGE, West Virginia University. A histological and histochemical study of the nasal gland of some non-marine birds. (15 min.)

Recent studies by Schmidt-Nielsen and others (Fed. Proc. 16: 113-114, 1957) and later studies demonstrating salt excretion by the large nasal glands of marine birds have led to an interest in their function in non-marine forms. The gland was also known to be relatively large in fresh water species such as dippers which suggests an adaptation other than for salt excretion in some aquatic species. A histological and histochemical study of previously preserved glands of the American Dipper *(Cinclus mexicanus)* was undertaken. For comparison, nasal glands as well as lacrimal, Harderian and various oral glands from the Turkey *(Meleagris gallopavo)*, Yellow-shafted Flicker *(Colaptes auratus)*, Rock Wren *(Salpinctes obsoletus)* and Wood Thrush *(Hylocichla mustelina)* were also examined. Variously fixed specimens (Bouin, formalin, Zenker-acetic acid, Zenker-formalin, Bensley, and Regaud reagents) were sectioned and stained with hematoxylin and eosin, mucicarmine, toluidine blue, periodic acid-Schiff reagent, and iron hematoxylin. With hematoxylin and eosin stain the tall, columnar cells of the nasal gland of *Cinclus* suggest mucous type cells in structure and staining properties, but periodic acid-Schiff reagent, mucicarmine, and toluidine blue revealed no mucous secretion. Nasal glands from the other species also gave negative reactions to these stains, but the oral, lacrimal and Harderian glands stained positive for mucin.

Although it may be concluded that the secretion of the nasal gland of these species is not mucoid, other secretion granules were not demonstrated and further study is needed to determine its nature. Possible functions of the nasal gland in non-marine birds are discussed and speculations about its evolutionary history are made.

85

CASIMER T. GRABOWSKI, University of Miami. Studies on the development of the notochord-forming center of the chick embryo. (15 min.)

What is the stimulus for the initial formation of notochord? It was postulated (Grabowski, '56) that the stimulus comes from the mesodermal cells themselves when they aggregate at some point in a critical density. Normally this is the node area. This hypothesis was tested in several ways. Studies of mesodermal cell density along the axis were made. Between stages 3 to 5 the cell density along the streak progressively increases, but it is always highest in the node region. Self-differentiating capacity was tested by grafting 0.5 mm squares of node mesoderm from DPS embryos, with or without overlying epiblast, to lateral regions of other DPS embryos. Grafts of pure mesoderm developed into notochord (20%) but did better if some epiblast was included (33-55%). Node area explants were also studied in a liquid tissue culture medium. Pure mesoderm could not be kept in a compact mass but would form either epithelial sheets or loose, vesicular masses. No notochord formation was observed in any of these cultures. Explants of whole nodes from DPS embryos (0.5 mm^2) formed notochords in 33% of cases. Mild versene treatment (50 mg % for 1 minute) completely inhibited notochord formation without affecting neural development. These experiments are not conclusive but do lend support to the critical density hypothesis. (Supported by a grant from the N.S.F.)

214

WILLIAM C. GRANT, Jr. and ALLYN J. WATERMAN, Williams College. Effect of light on thyroid activity in *Diemictylus viridescens*.

Waterman and Grant (1960) showed I^{131} thyroid uptake values for aquatic adults (scintillation counting) at $1.8 \pm 0.5\%$ of the injected dose and $15 \pm 7.2\%$ for terrestrial (eft) stages with plateauing at 72 and 120 hours, respectively. Recent investigations have shown uniformly low values between 0.01 and 0.09% for adults and efts kept on 12 hour day-night (diurnal) light cycles following two weeks of constant illumination. Animals in total darkness for two weeks following prolonged constant illumination showed increased uptake averaging 3% for adults and 16% for efts, values approximating earlier findings. Adults in total darkness for 32 days had uptake values of 12-19% and molted at frequent intervals, indicating increased thyroid activity.

Following several weeks exposure to diurnal light conditions adults kept in constant light, total darkness and 12 hour light cycles were studied at weekly intervals. Newts in constant illumination showed low values varying between 0.8-2.8% uptake. Both groups kept in darkness and under diurnal conditions showed initial high uptakes between 6-22% at the end of one week and then exhibited a leveling trend; after four weeks they approximated levels for animals under constant illumination. Although the interpretation of these results awaits further analysis it appears that illumination has a profound effect upon thyroid activity in the newt and that constant illumination depresses this activity. (Supported by grant A-1786 (C3) from the U.S.P.H.S., and grant G-7473 from the N.S.F.)

31

JOE L. GRIFFIN and PHILLIP R. SWEENY, Brown University. Physical and chemical studies on the external membranes of *Amoeba, Chaos* and *Pelomyxa*. (15 min.)

Previous observations indicate that at least 4 different layers make up the outer membranes of the carnivorous amebae, *Amoeba proteus* and *Chaos carolinensis (Chaos chaos, Pelomyxa carolinensis)*. The innermost layer sometimes separates from the microscopically visible membrane as it moves and seems to be a precipitate which forms from the hyaline cytoplasm at the membrane interface. Two other components, seen with the electron microscope, are a 70 A double membrane and, outside of this, a layer of fine filaments. The most external layer is microscopically invisible, but can be detected by observing adhering inert particles or by pressing the membranes of two cells together. Evidence indicates that narrow pinocytosis channels are filled with this invisible material. While inert particles adhere to the invisible layer, acidophilic particles apparently adhere to the filamentous layer and may cause adhesion of the membrane to underlying cytoplasm and formation of channels analogous to pinocytosis channels. Movements of the membrane as followed by neutral and acidophilic markers will be discussed. The membrane of the herbivorous ameba *Pelomyxa palustris* will be contrasted with those of the carnivorous amebae, from which it differs in the thickness of the outer layer and in other properties.

Previous workers have obtained histochemical evidence for carbohydrates in ameba membranes. We have found considerable carbohydrate in material prepared by alcohol precipitation of saline extracts of *A. proteus* and of slime from *Physarum*. Hexoses and hexosamines were identified in these alcohol precipitates by paper chromatography. These reults will be presented more fully and their significance discussed. (Supported by grant E-3410 from the U.S.P.H.S.)

186

LEONARD L. GROSSO, College of Saint Teresa. Peroxidase inhibition in fetal mouse thyroid. (10 min.)

C_3H mice weighing 25-30 g were used to obtain young. The adult animals were maintained on either a commercial or a purified diet containing 18% casein. Thiouracil, when used, was administered either by its inclusion in the drinking water or by incorporating it in the solid food. The animals were mated with normal non-treated males either before or after the start of thiouracil treatment. The mothers received the goitrogen for at least the latter two thirds of the ontogeny of the young. Young were sacrificed immediately after birth and their thyroids examined histologically and histochemically for peroxidase using the benzidine reagent. A number of thyroids were obtained from fetuses removed from the adult one day before the anticipated delivery date. The fetal or neo-natal organ was enlarged and hyperplastic with cells arranged in an afollicular mass. Little or no colloid was contained. The thyroid of control animals, obtained from non-treated mice, possessed follicles composed of normal epithelial cells and colloid. Histochemically peroxidase was found in the normal acinar epithelial cell. The enzyme was diminished greatly or was absent in the epithelial components of the young obtained from goitrogen-treated adults. (Supported by the Minnesota Division of the American Cancer Society.)

215

LEONARD L. GROSSO, College of Saint Teresa. Goitrogen-induced fetal liver alterations.

Reports concerning the effects of antithyroid chemicals on liver structure are quite contradictory. Extremes reported after the administration of these agents range from virtually no hepatic insult to a great structural alteration and even frank hepatic neoplasms. The present paper reports the changes produced in the livers of C_3H mice born of thiouracil-treated mothers and compares them with the normal organ. Thiouracil was administered by its inclusion in the drinking water or in the solid food, before or soon after her mating with a normal male. This timing means that the young, during all or most of their development, received goitrogen by way of the placenta. Livers were obtained from young sacrificed immediately after birth or one day before the anticipated delivery date. The large median lobe of the liver was examined histologically and the frequency of mitotic figures studied without the use of mitotic poisons. Although mitotic figures were present in the organ of mice from both normal and thiouracil-treated mothers, the number of divisions in all stages was much greater in the latter group. In addition a greater than normal number of multi-nucleate cells were present. Up to eight nuclei were found in exceedingly large cells. This indicates that karyokinesis without cytokinesis occurred a number of times. Many of the enlarged multi-nucleate cells had chromatin bridges between the nuclei. Sinusoids were enlarged and congested. (Supported by the Minnesota Division of The American Cancer Society.)

189

JACK E. HARCLERODE and ADAM ANTHONY, Pennsylvania State University. Thyroidal uptake and turnover of I^{131} by rats exposed to simulated altitude. (15 min.)

Thyroidal uptake and turnover of I^{131} were measured to determine the role of the thyroid gland in adaptation of rats continuously exposed to a simulated altitude of 20,000 to 23,000 feet. In view of indications that metabolic changes are most pronounced within the first few days of exposure, data were collected during the first seven days of exposure and for 15 to 22 days of exposure. Uptake of I^{131} occurred within three hours in both groups and showed no consistent difference between control and experimental groups. During the first two days of exposure the altitude-exposed animals showed a more rapid turnover of I^{131} as compared to control animals; however, after two days of exposure the turnover rate of the test group was comparable to that of controls. With longer periods of exposure (15 days) the differences in thyroid function were less marked.

The transient alterations in the thyroid gland were accompanied by changes in body weight, water balance and food consumption. These changes were most pronounced during the first few days of exposure, and represent the animals' metabolic adjustment to hypoxic conditions.

114

WERNER G. HEIM, Wayne State University. A reproduction-associated protein in the rat. (15 min.)

By means of vertical starch gel electrophoresis, a protein was demonstrated in the sera of pregnant, fetal and infant rats which is absent from the sera of adult male and adult, nonpregnant female rats. The protein has a mobility in the slow beta globulin range. It appears in the serum of the pregnant animal about the ninth or tenth day of gestation and persists during most or all the postnatal nursing period. It is present in the sera of the earliest fetal stages amenable to analysis, that is, at the fifteenth day of gestation. It persists throughout at least the first three weeks of postnatal life.

The relationship of this reproduction-associated protein with the various proteins reported previously in pregnant animals or in fetal animals, and with the growth-associated protein of Darcy (Brit. J. Cancer 11: 137, 1957), is presently under investigation. (Supported by grant G-10104 from the N.S.F.)

216

WERNER G. HEIM, Wayne State University. Quantitative relations between the fetal, neonatal and maternal serum proteins of the rat.

The sera of fetal and neonatal rats have been compared directly by paper electrophoresis in barbital buffer at pH 8.6 with the sera of the corresponding maternal animals. Determinations were made on 7 to 23 groups of animals daily from the 17th day of gestation through the second day postpartum and on the 7th and 24th to 30th post-natal days. Results show statistically significant correlations between the sera of the maternal animals and their offspring in regard to the relative concentrations of albumin, alpha-1, alpha-2 and gamma globulin, but not alpha-3 or beta globulin, for the antenatal stages and of albumin and gamma globulin for the postnatal stages. Similarly, significant correlations were found for the absolute concentrations of alpha-1 and alpha-2 globulin before, and for gamma globulin after, birth.

It is concluded that these results support, but do not prove, the possibility of the transfer of relatively large quantities of certain serum proteins from maternal to fetal or neonatal rat. (Supported by grant G-10104 from the N.S.F.)

217

L. G. HERSHBERGER, C. R. THOMPSON and P. M. ZAMBOANGA, Cutter Laboratories, Berkeley, California. Effect of carrageenin on cellulose sponge assay for anti-inflammatory substances.

Bush and Alexander have reported an improvement in the cotton granuloma assay for anti-inflammatory substances (Acta Endo. 35: 268, 1960). They found that the precision of the assay could be doubled by impregnating the cotton with carrageenin.

It was of interest to determine the effects of carrageenin in other foreign body granuloma assays for anti-inflammatory activity. Therefore, the effect of carregeenin on the cellulose sponge assay was studied. Plain cellulose sponges and sponges impregnated with 1% carrageenin were compared in 4 point bioassays of cortisol.

Addition of carrageenin increased control granuloma weights from 52.8 to 95.3 mg/rat. Slopes of the log dose-response curves for cortisol with plain and carregeenin sponges were – 15.0 and – 35.3, respectively. However, addition of carrageenin increased the standard deviation from the log dose-response regression from 7.27 to 14.1. Consequently the index of precision (λ = standard deviation/slope) of 0.40 for assay with carrageenin sponges was little better than the λ value of 0.48 for assay with plain sponges.

It was concluded that impregnation of sponges with carrageenin did not improve the precision of cellulose sponge granuloma assay for anti-inflammatory substances because both slope and standard deviation from the regression were increased.

218

K. K. HISAOKA and C. F. FIRLIT, Loyola University, Chicago. The distribution and localization of ribonucleic acid in the zebrafish embryo as determined by histochemical methods.

The distribution of ribonucleic acid (RNA) in the embryos of the zebrafish, *Brachydanio rerio*, from fertilization through post-hatching stages, was determined by using the methyl green-pyronin technique and ribonuclease-digested controls. RNA was distributed throughout the blastodisc and blastoderm but was concentrated most intensely along the free margin of the blastodisc and the epidermic stratum in the cleavage stages. During cleavage, the distribution of RNA in each blastomere remained relatively constant except for regional decreases during interphase and prophase. The spindle fibers and asters of all mitotic cells contained RNA. During the blastula stages, the distribution of RNA was uniform in all cells. As gastrulation was initiated, the highest concentration of RNA appeared first at the dorsal lip, then on the opposite side of the germ ring, as the ventral lip formed. Thereafter, the highest concentration of RNA was maintained in the presumptive nervous tissue (brain) and in the ventral lip as it continued to involute. The organs in order of the greatest RNA content were as follows: pancreas, digestive tract, liver, sense organs, brain, spinal cord, skeletal muscle and notochord. In the pancreas, the RNA concentration was uniformly high throughout development. The gastric mucosa initially contained little RNA but shortly thereafter, RNA synthesis increased to a maximal level at which it was maintained through the post-hatching stages. The other organs were characterized by the fact that RNA synthesis was initially high in the primordia and later declined to a low level as the organs became differentiated. (This investigation was aided by an institutional grant from the American Cancer Society.)

18

JACK R. HOFFERT and P. O. FROMM, Michigan State University. *In vitro* uptake of hexavalent chromium by RBC, liver and kidney tissue of the turtle. (15 min.)

Blood samples from the *Chrysemys picta* were taken by direct cannulation of the left aortic arch. The packed heparinized cells were washed three times with isotonic Ringer-phosphate-glucose buffer (pH 7.4). The uptake of chromium by cells was rapid initially, probably related to the amount of surface binding; this was followed by uptake at an exponential rate but without reaching an asymptotic value by 4 hours. The exponential uptake is related to the rate of diffusion through the cell membrane and to binding (either enzymatic or physical) to cytoplasmic proteins. Cell homogenates showed a linear uptake over a 4-hour incubation period. The binding of chromium to the cell surface or to the cytoplasmic proteins does not appear to be dependent upon metabolic activity since experiments in which NaF, NaCN, Na azide and $NaAsO_2$ (10^{-3}M) had no effect on uptake of Cr VI by RBC or slices of liver and kidney. Nucleated RBC when placed in 20 ppm chromium for ½ hr at 20°C show a significant decrease in their electrophoretic mobility. This indicates a reduction in the zeta potential due to binding of chromium at the cell surface. Fractionation and isolation of the RBC components has shown that chromium binds to the hemoglobin as well as to the cell ghosts and nuclei. (Supported by grant RG-6185 from the U.S.P.H.S.)

113

ROGER A. HOFFMAN, U. S. Army Chemical Research and Development Laboratories, Maryland. The reproductive system of male toadfish *(Opsanus tau)*. (15 min.)

Male toadfish were collected each month (May through January) from Chesapeake Bay. Gross and histological changes in morphology of reproductive tissues are described.

Testes are elongated organs lying along the dorsum of the body cavity, ventral to the kidney. They remain separate from each other along their entire length as do the spermatic ducts draining the testis tubules and coursing posteriorly through the pelvis to empty into the cloaca.

Spermatogenic cysts or inactive spermatogonia are frequently found along the posterior end of the spermatic ducts and can be found embedded on the medial edge of the accessory gland tissue. Thus, total castration would require removal of accessory glands and spermatic ducts as well as gonads.

Testis tubules are lined with spermatogenic cysts. Mitotic and meiotic divisions occur in these cells simultaneously and thus each cyst contains only cells of a specific stage of maturation. Six distinct stages of cellular morphology can be distinguished. At the appropriate time, the cyst wall ruptures and mature spermatozoa are released into the lumina. Phase microscopy reveals that these sperm have double tails.

Accessory glands of the male toadfish are large, fan-shaped organs, divisible into 4 regions, lying in the most posterior region of the abdominal cavity. Ducts which drain this glandular tissue join the spermatic ducts. At the posterior end, all accessory gland secretions and spermatozoa from each side are forced together through a pair of common ducts into the cloaca. (Supported in part by a grant from the Lalor Foundation.)

219

MARY J. HOGUE, University of Pennsylvania. The effect of poliomyelitis on guinea pig brain cells in tissue culture.

Guinea pigs do not contract poliomyelitis, however guinea pig brain cells in tissue cultures develop cytoplasmic changes and die when infected with types 1, 2 and 3 polio viruses. Nodules are often formed and sometimes extruded from the brain cells inoculated with any one of the 3 types of polio virus. The cells in the guinea pig body resist the action of the polio virus while the same types of cells in tissue culture are frequently destroyed by the virus. There is some variation in the time of appearance of the cytopathic reactions. Usually the cells infected with type 1 virus are the first to show changes, followed by those infected with type 3 and then type 2. The culture medium in which the virus was growing did not affect the tissue cultures. (Supported by grant B-1771 from the U.S.P.H.S.)

220

B. ELIZABETH HORNER, Smith College. Paternal care of the young and convulsive seizures in the grasshopper mouse.

Field and laboratory studies are in progress on *Onychomys torridus longicaudus* from western Nevada. The animals have been bred in captivity and males have been observed to assist in the care of the young. Following parturition the female appears more dominant than at other times. The male, normally fully as aggressive as the female, permits the female with newborn young to feed first and to appropriate the entire nest. Over a period of several days the male gradually works his way back into the main part of the nest and may then participate in washing and carrying the young. Instances have been noted, additionally, of behavior in which convulsive movements of the legs and head have been followed by periods of debility and stupor. From the age of four weeks to four months the animals appear especially vulnerable to such attacks, and in several cases the results have been fatal. (Supported by grant M-4096 (A) from the U.S.P.H.S.)

221

CARLA J. HUDDLESTON and LAWRENCE I. GILBERT, Northwestern University. Acid phosphatase activity in the testis of the Cynthia silkworm.

Past work has suggested correlations between acid phosphatase activity and protein synthesis, secretory processes, senescence, etc. During a general study of alterations in enzyme systems during insect metamorphosis, acid phosphatase activity of muscle, brain, fat body, blood, gut and gonad homogenates was determined spectrophotometrically using p-nitrophenyl phosphate as the substrate. The organs of choice proved to be the gonads, and par-

ticularly the testes since they do not break down during the pupal-adult transformation and can be extirpated with ease. The testis also showed the greatest activity in the diapausing pupa and the highest pH optimum (5.75). The rate of substrate hydrolysis by testes homogenates from developing animals was found to decrease to a level 50% of that of the diapausing pupa, and then to rise to the level of the diapausing pupa two days prior to ecdysis. Activity of the testis acid phosphatase is thus a mirror image of many other enzymes whose activity is at a low level during diapause but rises rapidly during development. Further studies revealed a general decrease of the pH optimum during adult development to pH 4.20 in the adult, suggesting that several acid phosphatases may be acting with peaks of activity at different developmental stages. Testes from pupal-adult intermediates obtained by juvenile hormone injection possessed acid phosphatase activity similar to that of developing pupae, but the pH optimum approximated that of the diapausing pupa indicating a "biochemical compromise" between pupa and adult in these endocrinologically disturbed animals. (Supported by grant A-2818 from the U.S.P.H.S.)

222

R. R. HUMPHREY, Indiana University. A chromosomal deletion in the Mexican axolotl *(Siredon mexicanum)* involving the nucleolar organizer and the gene for dark color.

White axolotls mated with dark ones commonly produce only dark offspring. Occasionally, however, one is white. These, if hypo-diploid (2N – 1), ordinarily have only one nucleolus, indicating that the nucleolar organizer and the gene for dark color are both in the missing chromosome. If the two are close together, the loss of only a short region instead of the entire chromosome could likewise eliminate both; this would account for those white offspring which are diploid but have only one nucleolus.

Recently a mating of white with dark produced a viable white diploid. Its cells contain two nucleoli, but one is much smaller than the other. This condition is best explained as resulting from deletion of a short segment of the chromosome containing the locus for dark color, this deletion either removing a portion of the nucleolar organizer or so affecting its action as to cause the nucleolus to be of subnormal size.

This unusual white, mated with an ordinary white diploid, produced F_1 about half of which have one of their two nucleoli small. Matings of two such F_1 produce offspring about one fourth of which have both the nucleoli of reduced size. These F_2, necessarily homozygous for the deletion, are of white phenotype and indistinguishable from their siblings. The white phenotype, usually regarded as resulting from mutation at the locus for dark, could therefore itself be the result of a deletion, but one shorter than that here reported and hence not affecting in like degree the nucleolar organizer. (Supported by grant G-4856 from the N.S.F. and grant RG-5850 from the U.S.P.H.S.)

223

PHILIP T. IVES, Amherst College. Some effects of Cobalt-60 γ radiation on premeiotic spermatogenic cells in *Drosophila melanogaster.*

Studies have been made on the induction of sex-linked lethal and autosomal dominant visible mutations, of translocations involving the Y and second and third chromosomes and of crossover and inversion recombinations, testing sperm from Oregon-R/*rucuca* which were deposited 9-16 days after irradiation by ♂♂ individually and exhaustively mated.

Induced crossovers were the only striking result, appearing in 129 of 15,326 flies after 2 kr and in 293 of 14,168 flies after 4 kr. Controls yielded 7 crossover flies in 67,010 from days 1-15 sperm. No inversions were recovered in tests of 609 non-crossover ♀♀ out of the 2 kr group and 547 from the 4 kr group. The crossovers were largely independent events in days 9-11 sperm and occurred mostly in small clusters in days 14-16 sperm, suggesting that late mitotic stages of spermatogenesis were the chief source of the days 9-11 sperm. There was no marked difference in total percentage of crossover flies in these two groups at each dosage.

Work in progress will test the frequency of associated recessive lethal mutations in crossover ♂♂ from days 9-10 and 15-16 sperm. (Supported by grant AT (30-1)-2467 from the U.S.A.E.C.)

224

SOPHIE JAKOWSKA, R. F. NIGRELLI and A. H. SPARROW, New York Aquarium and Brookhaven National Laboratory. Effects of chronic gamma irradiation on adult newts, *Diemictylus viridescens.*

Newts of both sexes were individually exposed to Co^{60} gamma irradiation at rates of 20, 40 and 80r/day for 1-6 weeks; to a total of 2100r with 2100, 300, 150 and 100r/day and to 1400-5600r at rates of 200-800r/day for one week.

Prolonged exposures were not lethal at lower rates (20-80r/day). Exposure to 2100r in one day caused death within 30 days; deaths were delayed 2-3 months when the same amount was administered at lower rates. All animals died within 30 days when exposed for 1 week to rates higher than 400r/day.

With the exception of occasional erythrocytic nuclear fragment (s) and class V-VI polymorphonuclear leukocytes, the blood picture was normal following exposure to lower rates. There was an increase in basophilic granulocytes immediately after the animals were exposed to 2100r in one day. Leukopenia, however, was persistent in all animals exposed to higher rates. Local phagocytic response occurred in animals challenged with bacteria, prior to expected death (29 days), after 7 days at 600r/day. Trypanosomes, when present, showed no morphological or cytological changes.

Histopathological examinations were made only following exposures to lower rates. The following changes were noted: (1) Spleen: progressive hemosiderosis, hyalinization, hemorrhages, thrombi, histiocytic infiltration; (2) Liver: pyknosis of hepatic nuclei, gradual disappearance of granulocytopoietic perihepatic layer and its replacement by a fibrous

capsule, hemorrhage and congestion; (3) Kidney: sloughing of tubular epithelium, cytoplasmic vacuolization and nuclear pyknosis, extensive hemorrhages; (4) Testis: nuclear pyknosis, crenation of interstitial cells, arrested spermatogenesis and necrotic changes. (A portion of this work was carried out at Brookhaven National Laboratory under the auspices of the U. S. Atomic Energy Commission.)

192

NORMA JOHNSON, Marquette University. Toxic effects of adrenal grafts to the chorioallantois. (Introduced by J. W. Saunders, Jr.) (15 min.)

Fragments (*ca.* 1 mm^3) of adrenal and of splenic tissue from adult fowl were placed on the chorioallantois of host chick embryos. Sham-operated controls and embryos receiving spleen grafts regularly survived to hatching, but hosts to adrenal grafts usually died within 24 hours after the operation. Frequency of the toxic effect varied with host age: for 9-day hosts the 24-hour mortality was 80% (53 of 66 cases) and for 13-day hosts, 53% (30 of 56 cases).

Toxicity to host embryos is likewise shown by grafts of embryonic adrenal glands and mortality increases with developmental age of the grafts and number of glands used. Thus, with 9-day hosts, graft of one adrenal gland from a 20-day embryo was lethal in 100% of cases, but graft of a single gland affected only 33% of hosts when 18-day donors were used and none when 13-day donors were employed. Mortality was 100% in 9-day hosts receiving 4 adrenal grafts from 18-day donors, 70% in those with two grafts and, as noted above, 33% in hosts which received only one graft. The graft of one 20-day adrenal, although providing less tissue than two glands from 18-day embryos, was toxic in a higher percentage of cases, indicating that factors determining toxicity of adrenal tissue are probably differentiating and increasing in amount as the donor approaches hatching. (Supported in part by a research grant to J. W. Saunders, Jr. from the N.S.F. (5006).)

107

JACK COLVARD JONES, University of Maryland. Observations on sexually depleted male *Aedes aegypti* (L.). (5 min.)

Aedes males can be artificially forced to copulate with a maximum of about 5 females but copulation may occur without insemination. After forced copulation with 5 females (3 inseminated) the seminal vesicles and accessory glands of the males do not differ from those of virgin males. However, when virgin males freely copulate with an excess of females for one hour, they inseminate about five, and their seminal vesicles, vas deferens and vas efferens are usually completely depleted of sperm, and their accessory glands reduced in diameter and contain little secretory material. The walls of the empty seminal vesicles are swollen and hyaline. The vas deferens in depleted males seem more contractile than in unmated controls. The testes of depleted males contain numerous undulating spermatozoa. The condition of the depleted males is maintained for at least 24 hours after being removed from the females. (Supported by grant RG-6021 from the U.S.P.H.S. Miscellaneous Publication Number 425, Contribution Number 3244 of the Maryland Agricultural Experiment Station, Department of Entomology.)

133

JACK COLVARD JONES, University of Maryland. Current concepts concerning insect hemocytes. (30 min.)

In the immature stages of many insects the number of hemocytes is believed to be maintained only by mitosis of circulating or sessile cells. In some insects a new set of hemocytes is formed during larval life from discrete imaginal discs and these cells are released before or during pupation. Nine morphologically distinct types of hemocytes have been described in a few insects, but only two or three of these types are universally present, and these universally present types are also the predominant hemocytes in insects possessing many other types. Insect hemocytes have been demonstrated to be phagocytic, to initiate coagulation of the hemolymph, to be involved in wound healing, to form tyrosine, to secrete tyrosinase, to store glycogen and to transport hormones. In some insects hemocytes are reported to transform into other tissues, principally into connective tissue and fat body. (Supported by grant H-5193 from the U.S.P.H.S. Scientific Article Number A 908, Contribution Number 3247 of the Maryland Agricultural Experiment Station, Department of Entomology.)

225

KLAUS D. KALLMAN, New York Zoological Society. Detection of gynogenesis in the viviparous teleost, *Mollienesia formosa* (Girard), by tissue transplantation.

The teleost, *M. formosa*, occurs abundantly in southern Texas and northeastern Mexico, but over its entire range it appears to exist primarily as females. In the laboratory and undoubtedly in nature, *M. formosa* females, when mated for many generations to the males of related species, produce exclusively female offspring that are phenotypically indistinguishable from the original *M. formosa* progenitor. It has been suggested by Hubbs that *M. formosa* reproduces by gynogenesis.

In order to test for the specific genetic relationship that has to exist between female parent and offspring in the case of gynogenesis, 11 *M. formosa* females (belonging to three different clones that had descended from three unrelated *M. formosa* females) were mated with *M. sphenops* and *M. latipinna* males and 375 tissue transplants were made from (a) the offspring to the parent, (b) among members of the same sib, (c) between members of different sibs but of the same clone and (d) between members of different clones. This transplantation test is based upon the fact that in vertebrates tissue transplants succeed only if all the donor's isoantigens are present in the cells of the host. Since in the case of nonsexual reproduction the offspring has inherited all its antigens and all other traits from the female parent, the offspring cannot possess any paternal antigens "foreign" to the female parent. Therefore transplants from gynogenetic offspring to the female parent must succeed.

All transplants were successful, if host and donor belonged to the same clone. If host and donor belonged to different clones, the grafts were rejected within one or two weeks.

Similarly, transplants from the offspring to the male parent were rapidly rejected. These transplantation experiments provide conclusive evidence that *M. formosa* reproduces by gynogenesis. (Supported by research fellowship HF-9500 to the author and by grant C-4945 to Dr. Sylvia Greenberg from the U.S.P.H.S.)

226

H. KALTER, Children's Hospital Research Foundation, Cincinnati. The teratogenic action of prednisolone on a genotype resistant to cortisone.

In the past it was found (Kalter, Genetics 39: 185, 1954) that F1 fetuses from the cross C57BL/6J ♀ × A/J ♂ are greatly resistant to the cleft palate (CP)-inducing action of cortisone acetate, there resulting 3.7% affected young, when maternal treatment was begun 11-1/3 days after conception. The present study was made to determine the response of such resistant fetuses to prednisolone acetate. Treatments and results were as follows: A treatment on each of 4 successive days of pregnancy beginning 11-1/3 days after conception of 2.5 mg hydrocortisone acetate, 2.2% CP, confirming the resistance found to cortisone (v.s.); and treatments on each of 3 or 4 successive days (denoted 3x or 4x) beginning 11-1/3 or 12-1/3 days after conception (denoted by 11-1/3 or 12-1/3, in parentheses) of 1.25 or 2.5 mg prednisolone acetate: 3 × 1.25 (12-1/3), 4.4%; 4 × 1.25 (11-1/3), 40.5%; 3 × 2.5 (12-1/3), 23.1%; and 4 × 2.5 (11-1/3), 92.8%. Finally, females given 4 × 0.0625 mg prednisolone/g body weight at conception (11-1/3) had 35.0% affected young. (Supported in part by grant RG-7107, U.S.P.H.S.)

45

FRED I. KAMEMOTO and SHARON M. KEISTER, University of Missouri. Cholinesterase activity in the crayfish kidney. (15 min.)

The association of cholinesterase with sodium transport has been reported by various investigators. Koch (In: Recent Developments in Cell Physiology, Academic Press, 1954) presented evidence for this association in the gills of the crab *Eriocheir sinensis*. He demonstrated the active uptake of sodium by the isolated gills, uptake being inhibited by the use of various cholinesterase inhibitors. Studies on the influence of cholinesterase in sodium regulation is being investigated in this laboratory.

Crayfish kidneys were homogenized in a glass homogenizer and cholinesterase activity determined by the titrimetric method with the Radiometer pH Stat at 25°C, optimum pH of 9.0 and optimum substrate concentration of 0.015 M AChBr. Cholinesterase activity (μM AChBr hydrolyzed/mg nitrogen/min) of kidney homogenates from animals held in tap water at 16°C was 0.27. Cholinesterase activities for the various parts of the kidney were as follows: coelomosac, 0.23; labyrinth, 0.14; tubule, 0.64; bladder, 1.10.

The injection of eserine into the hemocoel of crayfish causes an increase in the urine sodium concentration. If we accept the idea that the crayfish kidney is a filtration-reabsorption kidney and that cholinesterase is associated with sodium transport, we can now postulate that sodium is actively reabsorbed from the primary urine by the tubules and perhaps, also, by the bladder. The increase in urine sodium with eserine injections is probably due to the inhibition of cholinesterase in the kidney, preventing the reabsorption of sodium from the primary urine. (Supported by grant G-15990 from the N.S.F.)

227

HAROLD M. KAPLAN and D. J. O'BRIEN, Southern Illinois University. A comparative evaluation of the anesthetic properties of six barbiturates and of Viadril in the rabbit.

The common barbiturates and Viadril do not consistently induce deep surgical anesthesia in rabbits. The use of preanesthetics, or else supplementary maintenance with an inhalation anesthetic, as prescribed by others, is more reliable. The induction time for minimal anesthesia in sufficiently short except for Dial-Urethane and Veronal. The doses of barbiturates needed to produce deep anesthesia generally depress the respiration to a dangerous level. High doses of Dial-Urethane prolong anesthesia and the body temperature falls excessively. Rabbits vary considerably in their response to any given dose of each drug. The per cent of cases reaching deep anesthesia IV and IP, respectively, is: Kemithal 9.5, 9; Evipal 26, 11; Pentothal 20, 10; Nembutal 57, 25; Dial-Urethane 9, 42.8; Veronal 5.8, 0; and Viadril 23, 0. The duration of deep anesthesia is usually satisfactory with Kemithal IP, Pentothal IP, Nembutal IP or IV, Dial-Urethane IP or IV, and Viadril IV. The prolonged duration with Veronal favors the development of pulmonary complications. Nembutal is the most common barbiturate for rabbits, but at 35 mg/kg the average duration of deep anesthesia is only about 15 minutes. Higher doses increase the incidence of respiratory failure. Induction time is sufficiently short, overlapping the slow injection time. The animals remain in total anesthesia about 5 hours. There is too small a margin of safety between the anesthetic dose and respiratory failure.

51

FRANK F. KATZ, The Jefferson Medical College. Distribution of the nematode *Strongyloides ratti* in the small intestine of gonadectomized and sham operated male and female rats. (10 min.)

Sexually mature female Wistar rats were spayed or sham operated 10 days prior to receiving subcutaneously approximately 1150 filariform larvae in 0.1 ml of tap water. Male rats were castrated 10 days or sham operated 9 days prior to receiving the same inoculum. The inoculation process was such that no member of a group followed another of the same group.

The rats were starved 24 hours prior to being killed 7-14 days post-inoculation. The entire small intestine was removed and cut transversely into four approximately equal lengths. Each of these segments was opened longitudinally, cut transversely into 3 or 4 sections, and incubated in saline in conical flasks 4-7 hours at 37°C. After 24-48

hours refrigeration, the sediment was poured into Petri plates and covered with 20% glycerine in 70% ethyl alcohol. The alcohol was allowed to evaporate and the worms were counted directly under a stereoscopic microscope.

The total number of worms counted for 14 spayed females, 14 control females, 12 castrated males and 14 control males was 6773, 5464, 4827, and 5531, respectively. The distribution of the worms (in percent of total) in the first, second, third and fourth quarter of intestine, anterior to posterior, was as follows: spayed females, 81.5, 13.4, 3.1, 2.0; control females, 77.0, 16.3, 3.9, 2.9; castrated males, 82.8, 13.4, 2.3, 1.5; control males, 83.3, 13.9, 1.8, 1.0. No significant effect of surgical procedure or sex of the host was observed on the total number of worms recovered or on their distribution in the intestinal tract. (Supported by grant E-2894 from the U.S.P.H.S.)

30

NORMAN S. KERR, University of Minnesota. The inhibition of flagella formation by streptomycin. (15 min.)

When non-flagellated agar-grown cells of *Didymium nigripes, Physarella oblonga, Naegleria gruberi,* or *Chlamydomonas reinhardi* are suspended either in distilled water or M/200, pH 6.2 phosphate buffer they develop flagella within an hour or two. If streptomycin is added to the incubation medium, flagella formation is inhibited. The amount of inhibition is proportional to the concentration of streptomycin. Depending upon the organism under consideration, concentrations of streptomycin no greater than 10 to 100 microgram/ml are effective. If agar-grown cells of *Polytoma sp.* are suspended in a liquid growth medium together with various concentrations of streptomycin, flagella formation is inhibited. Cells of *D. nigripes* and *Polytoma* are able to multiply in the presence of a concentration of streptomycin sufficient to inhibit flagella formation. The effect of streptomycin on the growth of the other three organisms has not been determined.

If streptomycin is added to a suspension of cells which already possess flagella, the cells continue to be flagellated. If permanently flagellated or ciliated organisms such as *Astasia* or *Tetrahymena* are grown in the presence of 1 mg/ml streptomycin, their progeny continue to possess flagella or cilia. Streptomycin must therefore inhibit the differentiation of flagella rather than their retention or replication.

The effects of Cycloheximide and Chloramphenicol on flagella formation will also be discussed. (Supported by grant G-8776 from the N.S.F. and by a grant from the Graduate School, University of Minnesota.)

228

RICHARD G. KESSEL, Marine Biological Laboratory and Bowman Gray School of Medicine. Specialization of the free surface of ectoderm cells in planula larvae of *Aurelia aurita.*

Electron microscope observations were made on planula larvae fixed in 1% buffered osmium tetroxide. The free surface of the ectoderm cells possess numerous microvilli and cilia. The microvilli are long, filamentous extensions of the plasma membrane which interbranch forming a network around the cilia. The cilia possess an internal structure similar to that shown in a variety of other cells. The cilia contain nine peripheral rods, each of which appear double, and two central ones. The cilia terminate beneath the plasma membrane in a basal corpuscle similar to that described in the mollusc, *Elliptio* (Fawcett, 1958). Numerous fibrous "rootlets" arise from the lower pole of the basal corpuscle and extend downward into the cytoplasm. These rootlets are banded and possess an axial periodicity of 600-700 Å. Numerous vesicles are concentrated beneath the free surface of the plasma membrane and some appear continuous with the latter. Pinocytosis at the ultrastructural level appears to be a prominent mechanism for transport of material into and, perhaps, out of the larva. Numerous and varied inclusions located in both the ectoderm and endoderm cells will be described elsewhere. (Supported by grants RG-6942, B-2748 from the U.S.P.H.S.)

73

RICHARD G. KESSEL and NORMAN E. KEMP, Marine Biological Laboratory, Bowman Gray School of Medicine and University of Michigan. Electron microscope observations on the follicular envelope and developing oocyte in the tunicate, *Molgula manhattensis.* (15 min.)

Molgula oocytes become surrounded by inner and outer layers of follicle cells. Test cells migrate from inner follicular layer and embed in the oocyte. Inner follicle cells then become separated from periphery of oocyte by a chorion (vitelline membrane). Outer follicle cells are flattened, contain mitochondria and endoplasmic reticulum but do not appear active in secretion. Inner follicle cells enlarge during development and contain prominent cisternae of granular endoplasmic reticulum, mitochondria, Golgi elements, small vesicles, vacuoles of various sizes, and several large, lightly osmicated secretion granules. A characteristic inclusion develops in inner follicle cells and consists of electron-dense bodies embedded in a less dense matrix. This morphology probably results from fusion of dense bodies in vesicles of the Golgi region and material within endoplasmic reticular cisternae. Late in development, inner follicle cells fill with one or more large vacuoles in which the heterogeneous inclusions appear to degenerate.

Test cells contain prominent Golgi material, mitochondria and a system of often dilated cisternae of endoplasmic reticulum extending from nuclear to plasma membrane. Periphery of the nuclei of differentiated test cells consists of poorly osmicated material, metachromatic and PAS reactive, and is dotted externally by electron-dense spots which give it a porous appearance. Test cells eventually appear to degenerate and become incorporated into egg cytoplasm.

Plasma membranes of both oocytes and inner follicle cells contain microvillous projections into a filamentous chorion. The presence and distribution of numerous vesicles within oocyte suggest that micropinocytosis is a prominent mechanism for trans-

port of material between follicle cells and oocytes. Details of developing oocyte fine structure will be described. (Supported by grants (RG-6942C1, B-2748, RG-5867C2) from the U.S.P.H.S., and a grant (G-8773) from the N.S.F.)

78

J. F. KIDWELL, D. G. NASH, JANICE STADLER and J. W. GOWEN, Iowa State University. An X-ray induced deficiency in linkage group VI of the mouse. (15 min.)

A Balb B/Gw male, homozygous wild type at the belt locus, was exposed in the testis region to 500r of X-irradiation at 250 pkv, 30 ma with 0.25 mm Cu and 1 mm Al filtration. He was mated during the presterile period to a homozygous belt female. This mating produced a belt female of reduced body size. The hypothesis was formulated, and tested, that both effects were the result of a deficiency involving the belt locus. The small, belt female produced litters by homozygous belt and wild type males. All progeny from bt/bt males were belt and all from +/+ males were wild type. Approximately half the progeny, regardless of sire, were small. It was concluded that the belt locus (no new locus) is involved and that the size effect is dominant. Small, wild type females were mated to homozygous hairloss belt males. Approximately half the progeny were small. All the small progeny were hairloss belt and all the normal sized progeny were wild type. These results indicate a deficiency that includes both the hairloss and belt loci. Further tests indicate it does not include the naked locus. Small, wild type individuals were mated *inter se* to determine if the deficiency is homozygous lethal. Among a large number of progeny there was one extremely small male that was of the belt phenotype. It seems quite likely that this individual is homozygous for the deficiency. He is not hairloss. There are at least three possible explanations, assuming he is homozygous for the deficiency. (1) The belt mutation is, itself, a small deficiency and hairloss is not. (2) The physiology of the belt mutant is such that absence of the chromosomal material permits expression, while the material must be present in some form to permit phenotypic expression of the hairloss mutant. (3) Phenotypic expression of hairloss is dependent upon sexual maturity, and this male is not sexually mature. There has been some response to injection of growth hormone, but so far the treated male has not sired any progeny. (This work has received assistance from Contract AT (11-1)107 from the A.E.C. Journal Paper No. J-4115 of the Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa. Project No. 1180, 1187.)

66

JAMES R. KING and DONALD S. FARNER, Washington State University. The relationship of fat deposition and vernal migratory behavior as indicated by Zugunruhe in *Zonotrichia leucophrys gambelii*. (15 min.)

In many species of birds, the development of vernal migratory behavior in nature, or of *Zugunruhe* in cages, is preceded and accompanied by extensive accumulation of depot fat. In *Zonotrichia leucophrys gambelii* vernal fat deposition is the consequence of a marked, photoperiodically-induced hyperphagia. In experiments in which fat deposition in photoperiodically-stimulated birds is prevented by restriction of food intake there is neverthless the development of *Zugunruhe*. Also, *Zugunruhe* may develop *before* fat deposition when caged birds, under either natural or indoor conditions, are subjected to increased daily photoperiods in midwinter. In experiments in spring, the change from short to long daily photoperiods is followed *first* by fat deposition and then by *Zugunruhe*. This sequence is observed repeatedly in caged birds under outdoor conditions. Furthermore, fat deposition begins in wild birds *in advance* of vernal migration. From these data and observations it is obvious that neither *Zugunruhe* nor fat deposition is a direct functional consequence of the other. The only apparently tenable hypothesis is one of parallel photoperiodically-controlled timing mechanisms in which it is assumed that, although the receptors and some additional initial elements may be common to both, the final control pathways for these two phenomena are parallel and without direct functional relationships. This hypothesis can be extended to other photoperiodically-controlled phenomena in *Zonotrichia leucophrys gambelii*. Furthermore, it appears applicable to other migratory taxa of *Zonotrichia* and to other temperate-zone passerine migrants. (Supported by grant G-9872 from the N.S.F.)

165

JULIAN I. KITAY, University of Virginia School of Medicine. Sex differences in the effects of gonadectomy on pituitary and adrenal function in rats. (15 min.)

Male and female rats of the Sherman strain were gonadectomized at 21 to 30 days of age and studied 6 weeks later with intact animals used as controls. Increased pituitary weight was observed in both sexes associated with an elevation in pituitary ACTH content to 140% of the control level in castrated male animals. No change in pituitary ACTH was found in oophorectomized females. Adrenal weight was increased after gonadectomy in male rats and decreased in female rats. Peripheral plasma corticosterone concentrations at rest, after stress or after ACTH in operated males did not differ from those in similarly treated intact controls, whereas significant diminutions in plasma steroid levels were obtained in operated females. Gonadectomy shortened the biological half-life of corticosterone in male rats and prolonged it in female rats. Adrenal RNA and DNA content were both increased by castration in the males. A decrease in adrenal RNA with no change in DNA was observed in females. These findings suggest that castration is followed by increased ACTH synthesis and release in male rats. Adrenal secretory capacity is also enhanced, associated with adrenal hypertrophy and hyperplasia. On the other hand, both synthesis and release of ACTH are apparently diminished in female rats after oophorectomy. A corresponding diminution occurs in adrenal secretory capacity in association with adrenal atrophy without hypoplasia. (Supported by grant A-3370 from the U.S.P.H.S.)

229

J. B. KITZMILLER and W. L. FRENCH, University of Illinois. Chromosomes of *Anopheles quadrimaculatus*.

Preparations from the brain of fourth instar larvae and from pupal ovaries of *Anopheles quadrimaculatus* show two pairs of somatically paired mediocentric and one pair of subtelocentric chromosomes. Ovarian chromosomes measure 5.5, 8.0 and 8.25 microns. Testis preparations show one subtelocentric chromosome similar to that found in the ovary, but its homologue measures approximately 2.5 microns, suggesting dimorphic sex chromosomes. Salivary chromosomes with excellent, consistent banding patterns may be easily prepared by the aceto-orecin technique. The chromosomes average 67, 328 and 338 microns in length. The banding patterns show gross similarities to the patterns found in *Anopheles maculipennis* from Europe, but differ in details of banding. The x-chromosome is similar to, but not exactly like that of *A. messeae*. Maps of salivary chromosomes have been made. Puffs, Balbiani rings and interchromosomal and intrachromosomal connectives are present in the salivaries. Two types of polytene chromosomes, one condensed, the other attenuated, are found. Chromosomes of the Malphigian tubules do not show as distinct a banding pattern as the salivaries, but are quite usable in many ways. The Balbiani rings do not seem to be present and the puffs and connectives appear to show different localization in the Malphigian chromosomes. Six laboratory strains and one wild-caught strain all show a very similar heterozygous inversion and a long asynaptic region in chromosome III. More than 95% of individuals are heterozygous for the inversion in all strains examined. (Supported by grant E-3486 from the U.S.P.H.S.)

230

L. H. KLEINHOLZ, H. ESPER, C. JOHNSON and F. KIMBALL, Reed College and Marine Biological Laboratory, Woods Hole, Mass. Characterization and partial purification of crustacean eyestalk hormones.

Two activities of eyestalk extracts have been studied: (1) that effecting light-adaptation of the distal retinal pigment (DRPLH); (2) that causing increase in blood glucose concentration (HGH). Assay curves using dark-adapted *Palaemonetes vulgaris* have been standardized by measuring retinal response indices 45 minutes after extract injections. Extract containing DRPLH is largely inactivated within 12-24 hours at 20°C, but similar extracts, briefly heated at 100°C, retain their activity over this period. Eyestalk extracts from *Palaemon serratus* and from *Palaemonetes vulgaris* were incubated for varying intervals with trypsin and with chymotrypsin and were subsequently tested for DRPLH on *Palaemon xiphias* and *Palaemonetes vulgaris*, respectively. Extracts of *P. serratus* eyestalks incubated with chymotrypsin showed partial reduction, that incubated with trypsin showed nearly total reduction, in DRPLH activity. Almost complete loss of activity resulted from chymotrypsin treatment of *P. vulgaris* eyestalks compared with control extracts; trypsin treatment effected a reduction in but not a complete loss of activity. DRPLH can be dialyzed through cellophane membranes. These properties indicate the hormone may be a polypeptide.

Hyperglycemic hormone in eyestalk extracts of *Libinia, Pacifastacus* and *Pandalus* is inactivated by brief heating at 100°C. Unheated extract produces hyperglycemic responses roughly proportional to dosage of injected extract, but the dosage-response curves are not completely reproducible and require standardization. Hormonal activity is destroyed upon incubation with crystalline pepsin, trypsin and chymotrypsin. Partial purification of the hormone is obtained by fractional precipitation followed by column chromatography. (Supported by grants, G-3986 from the N.S.F. and B-2606 from the U.S.P.H.S.)

38

F. D. KLOPFER, Washington State University. Early experience and discrimination learning in swine. (15 min.)

Experiments were conducted to assess the effect of early training and mode of feeding on visual and spatial discrimination learning in swine. Position responses for food reward, learned as readily at 20 weeks of age as at 10, were unaffected by visual discrimination training.

When brightness discrimination was delayed until 20 weeks, neither feeder-fed nor floor-fed animals learned. When competition provided additional incentive, 5 of 30 animals showed some visual learning.

With early brightness discrimination training a large proportion of subjects learned, several of them successfully handling problems of wavelength, form discrimination and brightness reversals.

Swine have the capacity to utilize visual cues in obtaining food, but the ability requires special conditions and probably early training for its optimum development. If strong responses to non-visual food cues develop, vision will not later be used by the animal in this context.

100

ROBERT B. KLOPMAN, Cornell University. Nest building and reproductive behavior in *Agapornis* parrots. (15 min.)

The effects of nest building behavior on later reproductive behavior in the Lovebird, *Agapornis roseicollis*, were evaluated. Building activities considered were: cutting material, mandibulating it, tucking it in the plumage and carrying it to the nest-box. Treatments were designed to: (1) prevent the accumulation of a nest and (2) limit or prevent certain building activities. The effects of these treatments on courtship activities, copulation, egg-laying and incubation will be discussed. Results to date indicate clearly that preventing the accumulation of a nest and preventing all building activities except mandibulation have a differential effect on laying dates (Hinde and Warren, 1959), affect differences in clutch size but do not prevent egg-laying entirely. (This research was supported by N.I.H. Research Fellowship MF-8219.)

120

ELIZABETH A. KOCH and R. C. KING, Northwestern University. The origin of the vitelline membrane of *Drosophila melanogaster*. (15 min.)

The flexible, waterproof vitelline membrane lies between the chorion and the plasma membrane of the oocyte. Cytochemical studies have shown that the vitelline membrane contains proteins, polysaccharides and lipids. During their formation the plasma membranes of both oocyte and follicle cells are thrown into numerous interdigitating microvilli, and the vitelline membrane is produced by the coalescence of numerous vitelline bodies which accumulate in the intercellular space. Subsequently the chorion is laid down by the follicle cells upon their basement membranes. The question naturally arises as to whether the follicle cell or the oocyte plays the more active role in the secretion of the vitelline membrane. Studies of abnormal oogenesis in females homozygous for either of two recessive, female sterile genes provide information on this point. The mutants are *tiny* (*ty*, 1-44.5) and *female sterile* (*fes*, 2-5 $\pm$). In the case of *ty*, rare instances were observed where vitelline membrane was secreted by columnar follicle cells against the plasma memebrane of a nurse cell. In *fes* ovaries vitelline membrane is sometimes secreted by columnar follicle cells against the plasma membranes of myriads of undifferentiated tumor cells. It thus appears that once the follicle cell reaches a certain degree of maturity is secretes precursor material of the vitelline membrane into the intercellular space. Normally the plasma membrane of the oocyte forms the opposite border of the intercellular space; but a plasma membrane supplied by some other cell may serve this purpose. (Supported by grant G-11710 from the N.S.F.)

128

I. L. KOSIN and S. H. WEISBROTH, Washington State University. "Physiological zero" of turkey blastoderms and genetic differences in their mitotic index. (15 min.)

If "physiological zero" in turkey early embryonic development is defined as the temperature below which no visible signs of cellular activity can be detected, then that point in the domestic turkey is below 13-14°C. Furthermore, in the same organism, a hereditary predisposition to high hatchability is reflected by a higher mitotic index than in the case of a hereditary predisposition to rapid post-hatching somatic growth. Evidence to this effect has come from a cytological study of Broad Breasted Bronze eggs from two genetically distinct lines. The latter have evolved through a six-generation, single-trait selection plan: Line 2 has been selected for large adult body size, Line 4, for high hatchability. Mitotic Index (MI), as revealed by the Feulgen technique, and based on 500 cells for each of 10-15 blastoderms per treatment, per line, was the criterion of visible cellular activity. Also, the incidence of "necrotic" mitotic figures (obtained on the same basis and from the same experimental material as MI) was determined. Eggs stored for 24 hours at 13-14°C before fixation (treatment 10) showed MI of 1.27 and 1.74 for lines 2 and 4, respectively. MI of eggs fixed immediately after oviposition (treatment 00) were 2.26 and 3.85. Eggs stored at 13-14°C for 14 days and then incubated for 5 hours prior to fixation (treatment 21) yielded MI of 2.88 and 3.33. The incidence of "necrotic" mitoses, compared by lines and treatments, was: treatment 10, line 2—0.73, line 4—0.74; treatment 21, line 2—1.87, line 4—1.96. (Supported, in part, by grant RG-5544 from the U.S.P.H.S.)

49

CHARLES E. LANE, University of Miami. Observations on the general biology of *Physalia*. (15 min.)

Gas secretion, synthesis of a peptide toxin and nutrition of remote tentacle structures in the siphonophore *Physalia physalis* are shown to depend on the existence and circulation of a specific gastrovascular fluid. While this liquid is isotonic with sea water it differs qualitatively from sea water in important ways. It may contain as much as 1 g/liter of protein, the peptide and amino acid composition of which appears to be identical from animal to animal regardless of age, season or dietary state. The tubular system within which this fluid circulates and the effective propulsive mechanims will be described. Finally, the relationships between gastrovascular fluid, toxin synthesis and gas secretion will be discussed. (Supported in part by contract #NONR 840 (17) between O.N.R. and the University of Miami.)

46

JAMES L. LARIMER, University of Texas. Gas secretion and respiratory physiology of the Portuguese man-of-war, *Physalia*. (15 min.)

The float gases of *Physalia* have been shown to contain carbon monoxide in addition to atmospheric gases (Wittenberg, J. Exper. Biol. 37: 698, 1960). The CO originates from the metabolism of serine in the gas gland *(ibid.)*.

Complete analysis of the gases of a large number of individuals using both gas chromatography and mass spectrometry indicates large variation in composition; however, all animals were found to contain CO. Evidence will be presented which indicates that CO is the only gas produced by the animal, and that the inflation of the float by a mixture of gases results from the diffusion of N_2, O_2, and A into the gas space from the atmosphere and surrounding sea water.

The animals could not be induced to secrete when the float was deflated; however, gas replacement techniques allowed an estimated rate of CO secretion to be obtained. These rates were found to vary from about 20 to 150 μl/hr per animal.

Data will be presented to indicate that the tissue respiration of the animal is uninhibited by 70% CO, yet both CN^- and N_3^- were found to inhibit respiration strongly. Attempts to demonstrate cytochromes by difference spectra after oxidation and reduction were not successful. The terminal O_2 acceptor is still unknown at the present time. (Supported by grant 13318 from the N.S.F.)

138

J. LASH, F. ZILLIKEN and F. A. HOMMES, University of Pennsylvania and R. K. Universiteit,

the Netherlands. Chondrogenic induction with a low molecular weight tissue component. (15 min.)

Explanted somites only form significant quantities of vertebral cartilage if exposed to the inductive influence of the embryonic spinal cord or notochord. This reaction is specific in the sense that no other tissues have been found to induce this response (cf. Holtzer, '61, review). Extracting with cold perchloric acid, a partially purified nucleotide-containing component has been obtained from the embryonic spinal cord and notochord that simulates the chondrogenic action of the intact tissues. Out of 228 cultures of chick somite clusters, 186 have been induced to form cartilage by adding nucleotide fractions. In contrast, 304 control cultures yielded 38 small nodules of cartilage. Although extracts of the notochord or the ventral half of the spinal cord induced cartilage formation, extracts of the dorsal half of the spinal cord had no effect upon the somites. Moreover, nucleotide fractions of other embryonic tissues (epidermis, endodermis, muscle, limb buds) did not induce cartilage formation.

Preliminary experiments on the characterization of the active component in the nucleotide fraction indicate that it is of relative small molecular weight (MW 8000-10,000) and is probably an oligonucleotide. (Supported by grant A-1980 (C2) from the U.S.P.H.S. JL: Helen Hay Whitney Established Investigator; FAH: Postdoctoral Fellow, R.K. Universiteit. Nijmegen.)

156

ROBERT C. LASIEWSKI, University of Michigan. Energy metabolism of hummingbirds. (Introduced by J. M. Allen) (15 min.)

Effects of temperature on energy metabolism of torpid, resting and active hummingbirds were determined with 24 birds representing four species (*Calypte costae, C. anna, Archilochus alexandri* and *Selasphorus rufus*). They ranged in weight from 2.6 to 4.6 g. Metabolic rates of torpid hummingbirds varied exponentially with ambient temperature, rising from 0.17 cc $O_2/g \times hr$ at 15°C to 1.95 cc $O_2/g \times hr$ at 32°C. Body temperatures of these birds were generally within 1°C of the environment. It was possible to maintain postabsorptive birds resting in the dark at various temperatures in a non-torpid state long enough to obtain data permitting construction of a metabolism-temperature curve comparable to those characteristic of obligate homeotherms. The lower critical temperature from this curve is 28.5°C and the metabolism within the zone of thermal neutrality ranges from 3.0 to 3.5 cc $O_2/g \times hr$. These metabolic values are lower than the ones predicted for birds in this weight range. This seems to be associated with the fact that hummingbirds regulate body temperature at a level 5° or 6°C below that characteristic of activity, when they are maintained in the dark. Below thermal neutrality, metabolism of resting birds rises with decreasing ambient temperature at the rate of 0.54 cc O_2 $(g \times hr \times °C)^{-1}$. The best line which can be fitted to the data extrapolates to zero metabolism at 35°C, which is consistent with the observations on body temperatures of birds resting in the dark.

The range of metabolism of hummingbirds in the light is impressive, extending from 6.7 to 45.7 cc $O_2/g \times hr$ depending on ambient temperature and level of activity. A metabolism-temperature curve such as that characterizing obligate homeotherms can be fitted to the lowest levels of metabolism observed at various temperatures in the light. A value of 6.7 cc $O_2/g \times hr$ is the lowest found in the zone of thermal neutrality. Hummingbirds do not necessarily become torpid at night, and can maintain homeothermy over a wide range of ambient temperatures. (N.S.F. Cooperative Graduate Fellow. Research supported in part by grant G-9238 from the N.S.F. to W. R. Dawson.)

231

HOMER B. LATIMER, PAUL GIBBONS ROOFE and LILY S. FENG, University of Kansas and Jackson Hole Biological Station. Weights and measurements of the body and of some of the organs of the tiger salamander.

The body weight, weights of 17 organs, body length and 13 additional linear measurements were made on 42 *Amblystoma tigrinum melanosticum* from three separate ponds in the Grand Teton National Forest, Wyoming. Fifteen organ weights were determined for 24 *Amblystoma tigrinum* from Minnesota. The body weight, body length and sex of the Minnesota specimens are not available.

Specimens from two of the Wyoming ponds are not significantly different quantitatively and there are no significant sex differences in any of the groups. The salamanders from one Wyoming pond are heavier, longer and broader bodied. The organ weights as percentages of body weight, however, are in many cases greater in the smaller salamanders.

The weights are quite variable, with the organ weights of the Minnesota specimens the least variable. The coefficients of variation are about the same for the weights in grams and for the percentage weights.

The linear measurements are less variable than the weights and also much alike for the measurements in millimeters and as percentages of body length.

The Wyoming salamanders have smaller eyes, heavier lungs and heavier and longer digestive tracts than the Minnesota specimens.

The organ weights are poorly correlated with body weight but the external measurements are well correlated with body length in the Wyoming Amblystoma. (Supported in part by grant from the Department of Health, Education and Welfare, National Institute of Health B-113 (C9).)

232

MALCOLM S. LEA and LAWRENCE I. GILBERT, Northwestern University. Cell division in diapausing silkworm pupae.

Some of the physiological effects of wounding diapausing silkworm pupae resemble events which normally follow the secretion of ecdysone (insect molting hormone) at the initial stage of development. These include quantitative changes in res-

piratory rate, cytochromes, blood and tissue proteins, etc. A phenomenon common to all developing insects is cell division, but this has not been directly demonstrated in diapausing pupae although Davis & Schneiderman (1960) found increased thymidine uptake by cells of the epidermis and blood after severe wounding. While investigating the physiology of insect hemocytes we have observed mitotic activity in the blood cells of diapausing *Hyalophora cecropia* and *Samia cynthia* pupae. Diapausing *Samia cynthia* pupae were wounded and the blood cells observed *in vitro* by phase contrast microscopy and in Bouins-fixed, hematoxylin-stained (Harris) smears. No mitosis was detected in blood removed from an initial small puncture, but dividing cells of the proleucocyte class were noted within 18 hours. 20-30 minutes were required for anaphase and telophase.) Aseptic puncture without blood loss, and destruction of epidermal cells by surface cautery were similarly effective. The response can be elicited in brainless pupae, and isolated abdomens prepared from diapausing pupae reveal hemocyte division more than 1 month after the operation. An increase in blood cell count was noted (*e.g.* from about 800 to 7,000 cells/mm³ in response to a series of wounds). Whether this is due entirely to division of circulating cells or additionally to enhanced activity of a hemopoietic tissue and/or release of cells adherent to other tissues is being investigated. (Supported by grant A-2818 from the U.S.P.H.S.)

123

NAOMI LEMKEY and HISAKO O. YOKOYAMA, Medical School, Northwestern University. Esterases and phosphatases of the developing and adult mouse brain. (15 min.)

The zymogram method of Hunter and Markert (Science, 125: 1294-5, 1957) for the separation and identification of tissue enzymes on starch gel was applied to the study of the mouse brain. The success of this method depends to a great extent on the amount of enzymes extracted and maintained in a reactive state, and their net charges which affect the electrophoretic mobility. Organic solvents, anionic and non-ionic detergents, and various salts have been compared as to their suitability for preparing brain homogenates. The esterases extracted by these methods were separated electrophoretically and tested for their ability to hydrolyze some substrates used for histochemistry: α naphthyl acetate, naphthol-AS-acetate, α naphthyl butyrate, 5-bromoindoxyl acetate, acetylthiocholine and thiolacetic acid. Inhibitors and activators were also used to further characterize these esterases.

Preliminary trials have been made to separate electrophoretically the esterases and phosphatases at successive stages of the developing brain. Foetal brains from 11 to 19 days were studied and the electrophoretic enzyme patterns noted in relation to the patterns in neonatal and young adult brains. The acid phosphatases and esterases show patterns varying with the stage of development while the alkaline phasphatase is characterized by a single migrating enzyme band in foetal and very young neonatal brains. (Supported by Predoctoral Fellowship MF-10,961 (C1), U.S.P.H.S.)

233

JOHN F. LHOTKA, Jr. and H. A. DAVENPORT, University of Oklahoma and Northwestern University Medical Schools. Congenital polycystic kidney in the black hooded rat.

An attempt was made at Northwestern University Medical School between 1947 and 1950 to develop a strain of hooded rats which would be resistant to the respiratory infections so common to animal houses. Both black and yellow hooded animals were interbred and by the project's termination, over 1000 rats had passed through the colony. Early in the study, it was noted that one strain of black hooded rats possessed heredity for congenital polycystic kidney. To complement the paper by Morgan (Anat. Rec. 115, 635), we believe that a brief report on these animals will be of value. This polycystic strain was followed through 7 direct generations, as well as through cross-breeding studies with normal rats, and totaled about 100 animals. The condition was recessive and was restricted to the kidneys. No genital anomalies or involvement of other internal organs were noted. Both sexes were equally susceptible. The polycystic anomaly was bilateral although sometimes one kidney would be more involved than the other. The condition was evident as early as the 4th postnatal week by necropsy but clinical signs of renal involvement did not appear until about the 5th to 6th month. All animals, unless they previously succumbed to respiratory infections (to which they were increasingly susceptible with age) or had been sacrificed, died in uremia between 6 and 12 months, about 9 months being the average lifespan. On necropsy, the kidneys were usually twice normal size and presented the typical histopathological picture of polycystic disease.

155

PAUL LICHT, University of Michigan. Effects of temperature on heart rates of lizards during rest and activity. (Introduced by W. R. Dawson) (15 min.)

Information available on physiological responses of lizards to temperature deals primarily with resting individuals, and no quantitative studies of the influence of temperature on the capacities of these animals for activity have been reported. In an initial attempt to estimate such capacities, studies were made of the increment between resting and maximum heart rates at various temperatures in the lizards *Dipsosaurus dorsalis, Uma notata, Sceloporus graciosus* and *Tiliqua rugosa*. This increment was used as a preliminary index of cardiac reserve.

Heart rates were determined from electrocardiographic records. Resting values were obtained from animals after they had remained quiescent for at least an hour at a particular experimental temperature. Maximum values were taken as the highest that could be produced in these same animals with prolonged, vigorous mechanical stimulation. Within the thermal limits for each species, both resting and maximum heart rates vary directly with temperature, the maximum rates ranging from double to triple the resting rates. The curve for maximum rate diverges progressively from that for

resting rate up to a temperature ($T_{A\ max}$) 5-10°C below the lethal level for the species. Above the $T_{A\ max}$ the slope of the curve for maximum rate is less steep, producing a reduction in the increment between the two curves. Heart rate during spontaneous activity rarely reached the level associated with stimulation except at temperatures exceeding the $T_{A\ max}$. While the $T_{A\ max's}$ obtained for the four species—*Dipsosaurus*, 42°C; *Uma*, 39°C; *Sceloporus*, 37°C; and *Tiliqua*, 32°C—were, with the exception of that for *Tiliqua*, 1-3°C above their respective preferenda, they correlate well with the extent of the heat resistance evident in these animals. (Supported in part by grant G-9238 from the N.S.F. to W. R. Dawson.)

234

V. F. LINDEMAN, Syracuse University. Phosphatases in the rat testis following treatment with Diethylstilbestrol.

Diethylstilbestrol when administered to mammals results in the inhibition of growth and differentiation of the germinal cells of the testis. Extended treatment may result in complete depravation of the cells of the seminiferous tubules. In the present study phosphatase activity of rat testis was studied during extended administration of this estrogenic agent. Alkaline and acid phosphatase, 5′Nucleotidase, and Teppase (Lindeman and Rosenbaum, Anat. Rec. 132, 469) were studied. Rats administered 0.5 mg Diethylstilbestrol twice weekly were sacrificed at 5-day intervals, testis removed, weighed, homogenized, and incubated with substrates. Activity is stated in micrograms P hydrolyzed from substrate per gram of tissue per minute.

Alkaline phosphatase activity did not change from normal (40) for 55 days after which it decreased to half at 100 days. Acid phosphatase remained normal (80) for 35 days after which it decreased to half by 100 days. 5′Nucleotidase activity began to increase after 10 days (normal 305) in a linear order for 55 days (maximum 685). No further change to 100 days. Teppase activity began to decrease (normal 36-40) after 10 days to 6-8 at 50 days.

Since Teppase is present only in germinal cells of the testis (above communication) this enzyme reflects the depravation of the cells in the seminiferous tubules. Therefore the increase in the 5′Nucleotidase activity appears to be associated with cells other than the germinal tissue. Histological preparations indicate an increase in connective tissue formation.

235

V. F. LINDEMAN and FREDERIC GREENBERG, Syracuse University. The effect of feeding Furadroxyl upon the 5′nucleotidase activity of the rat testis.

There is ample evidence in the literature that feeding nitrofurans to rats results in the arrest of spermatogenesis. In this study rats were fed 1.5 g per kg per day Furadroxyl. Animals were sacrificed at intervals of from 3-8 days, their testis removed, weighed, homogenized and incubated with adenosine 5′ phosphoric acid. The activity of the phosphatase, 5′nucleotidase, was calculated as the number of micrograms of P hydrolyzed from the substrate per minute per gram of tissue at 37.5°C.

The 5′nucleotidase activity of mature rat testis normally measures approximately 300 micrograms. This series averaged 295. The first appreciable change in enzyme activity was detected after 7 days of feeding. At this time the activity measured 340. The following values were obtained on succeeding days: 9 days, 420; 12 days, 485; 15 days, 520; 21 days, 580; 30 days, 630; 38 days, 660. Removal of the drug from the food at this time results in the gradual return of the enzyme to normal within 60 days. Histological preparations reveal that cytological changes leading to almost complete depravation of the germinal elements in the seminiferous tubules, parallels the increase in the enzyme activity. Restoration of spermatogenesis following cessation of drug feeding results in the return to normal enzyme activity. It appears that this enzyme might be associated with some function in the testis other than spermatogenesis.

236

WILBUR M. LUCE, University of Illinois. The effect of lactamide upon the facet number in the mutants forked Bar, forked double Bar, forked Infrabar-Bar and forked wildtype in *Drosophila melanogaster*.

Previous work mainly by Dr. M. Ogaki has shown that lactamide when fed, in sublethal concentrations, to larvae at definite times markedly increases the facet number of the Mutant Bar in *Drosophila melanogaster*. The study reported here concerns similar experiments with forked Bar and several other related mutants of the Bar group. All the mutants carried forked (f) a bristle mutant used to help detect contamination. The results with forked Bar (fB), in agreement with M. Ogaki, show very pronounced increases. Thus, larvae, raised at 25 ± 0.2°C, when transferred to food containing 3% lactamide when they were 67-73 hours old, reckoning from the time of deposition as eggs, had when they emerged as adults in females a mean facet number of 554 ± 1.3 and in males a mean of 552 ± 8.5 as compared with controls of 76 ± 0.7 for females and 84 ± 0.3 for males. Forked wildtype (fB+) untreated when raised at the same temperature had a mean facet number of 731 ± 2.3 for females and 639 + 3.6 for males. Forked wildtype (fB+) males treated with lactamide (3%) gave a mean facet number of 651 ± 13.0 showing that this chemical does not increase facet number in normal or wildtype eye. Significant but not as striking increases as in Bar were found when forked double Bar (fBB) and forked Infrabar-Bar (fB^1B) were treated with lactamide (3%). Thus, for example, forked Infrabar-Bar females treated with lactamide (3%) had a mean facet number of 148 ± 12 as compared with the untreated controls of 38 ± 1.0. Similar results were obtained with forked double Bar. (The experimental part of this work was carried out in the Division of Biology at the California Institute of Technology.)

168

J. G. MACKIN, Agricultural and Mechanical College of Texas. Oyster leucocytes in infectious disease. (30 min.)

Leucocytes of oysters function (1) in phagocytation and subsequent diapedesis to remove infectious fungi, protozoa and bacteria, and (2) to isolate infected centers with massed leucocytes which later form epithelial tissue surrounding the zones of infection. In this type of defense mechanism, the entire infected area may be discharged through the external epithelium. The capacity of leucocytes to form epithelial tissue is also demonstrated by the isolation of mud globules injected into the adductor muscle. The steps in isolation consist of (1) removal of tissue debris by phagocytation, (2) isolation by masses of unorganized leucocytes, (3) consolidation of leucocytes to form a secreting epithelium, (4) secretion of conchyolin and subsequently of nacre and (5) migration of muscle to new part of the shell thus transferring the walled-off nodule of mud to the mantle area.

Some reactions of oyster leucocytes to infection are so concentrated that the reaction may result in death of the host. Massive infiltration of the stomach epithelium may cause rupture of the mass into the gut lumen, carrying the epithelium with it. Oysters so affected die apparently from the mechanical effects of massive reaction.

Some few disease-producing organisms apparently fail to produce cellular reaction. In one such, about 80% of infected oysters recover without development of cellular reaction, indicating that antibodies may be produced.

176

ROBERT GILMORE McKINNELL, Institute for Cancer Research, Philadelphia. Intraspecific nuclear transplantation in *Rana pipiens*. (15 min.)

Nucleo-cytoplasmic hybrids have been made using *Rana pipiens* (Kandiyohi dominant mutant) and wild-type *R. pipiens*. The study was designed to answer the following questions: Is a nucleus derived from a mutant donor competent to order egg cytoplasm from a wild-type donor to develop in such a manner that the mutant gene is expressed in the resulting offspring (preliminary results have been published elsewhere (McKinnell, Amer. Nat. XCIV: 187-188, 1960))? Does the reciprocal transplantation yield comparable results, *i.e.*, is the wild-type phenotype obtained when nuclei from wild-type donors are transplanted to egg cytoplasm derived from a mutant donor?

Animal hemisphere blastula nuclei from normally fertilized eggs were transplanted to activated enucleated eggs. Wild-type *R. pipiens* were obtained from Vermont and *R. pipiens* (Kandiyohi dominant mutant) were obtained from Wisconsin. Results of this study indicate that (a) conventional transplantation procedure is feasible using eggs of the Kandiyohi mutant frog, (b) the geographical hybrid of Vermont egg cytoplasm and Wisconsin diploid nucleus, and the reciprocal cross, is compatible and may develop into juvenile adults and (c) with reference to wild-type and Kandiyohi nucleo-cytoplasmic hybrids, the genic pattern of the transplanted nucleus is expressed in successfully developed recipients. (Supported by grant E 71 C from the American Cancer Society.)

121

ANTHONY P. MAHOWALD, The Johns Hopkins University. Electron microscopy of early embryogenesis in *Drosophila melanogaster*. (Introduced by T. R. F. Wright) (15 min.)

The changes in fine structure of the *Drosophila* embryo have been followed during early embryogenesis. A search was undertaken for evidences of mosaicism but with the exception of the pole cells no distinguishable regions were found. The pole cells have, besides the polar granules with a myeloid-type structure, a distinctive nucleolus and endoplasmic reticulum.

Special emphasis has been given to the transition period between the syncytial blastema and cellular blastoderm stages. The new cell membranes form from the inward growth of one of the multiple folds in the plasma membrane. These folds only occur in the regions between the nuclei of the blastema. At the same time as the membranes are moving inward the nuclei are elongating in a radial direction. Many small vesicles become associated with the tip of the furrow, but their origin and fate remains doubtful. It is suggested that their origin may be from the nuclear membrane. At later stages continuities have been observed between the nuclear and cell membranes. The endoplasmic reticulum is composed primarily of large lamellae with some ribosomes attached to the membranes. The strong basophilia of the cytoplasm is due to the large number of unattached ribosomes. (Supported by a Predoctoral Fellowship from N.S.F.)

116

THOMAS E. MALONE and K. KENNETH HISAOKA, Loyola University, Chicago. The formation of deutoplasmic components in the zebrafish ovary. (15 min.)

The development of deutoplasmic inclusions in oocytes of the zebrafish, *Brachydario rerio*, was studied using histochemical techniques for lipids, carbohydrates and proteins. Morphological correlations were made with cytological methods for mitochondria. This approach has resulted in the identification of two types of yolk spherules which differ both in size and in histochemical characteristics. One type, designated as extravesicular yolk, first appears in young oocytes as small sudanophilic granules about 0.5 μ in diameter. These granules also contain carbohydrate and protein moieties. As the oocyte grows, these yolk granules increase in size and reach a maximum diameter of about 15 μ in the mature oocyte, with no significant changes in histochemical properties. The extravesicular yolk granules appear to be derived from mitochondria. The second type of lipid inclusion, designated as intravesicular yolk, originates in young oocytes as sudanophobic vesicles about 3 μ in diameter. These appear as vacuolated spaces with Sudan Black B staining. These vesicles are not empty for they stain intensely with the PAS reaction for carbohydrates. With Bromphenol Blue, they are protein-negative. In oocytes in intermedi-

ate stages of cytoplasmic differentiation, the intravesicular spherules increase to a maximum diameter of about 31 μ and are still sudanophobic and PAS-positive but show a faint, central accumulation of protein substance. Just prior to full cytoplasmic differentiation, the majority of these intravesicular spherules become sudanophilic, rich in protein and, at the same time, negative to the PAS reaction. The relationship of intravesicular yolk to cytoplasmic organelles is not clear. (Supported by an institutional grant from American Cancer Society.)

41

KUNIO MATSUMOTO, Okayama University and University of Oregon. Peripheral secretion of neurosecretory cells in ventral ganglia of decapod crustaceans. (Introduced by B. T. Scheer) (15 min.)

The eleven types of neurosecretory cells previously demonstrated in Japanese crabs (Matsumoto, Biol. J. Okayama Univ. 4 103-176, 1958) can be seen in the crab *Hemigrapsus nudus* (Dana) and the crayfish *Pacifastacus leniusculus* (Dana). The large A cells (70-90μ), seen in the brain and ventral ganglia, secrete two products: acidophile granules, transported in the axon, and basophile granules, discharged peripherally. The cycle of peripheral discharge will be demonstrated. Small B′ cells (30-40μ) occur only in the ventral ganglia and secrete basophile granules peripherally.

237

RAOUL MICHEL MAY and FRANCOISE LAHILLE, Faculté des Sciences, University of Paris. Direct action of X-rays on the spleen of chick embryos during its myelogenesis.

In order to study the effect of X-rays during the myelogenic period of the chick spleen, we used embryos on the 11th day of incubation and doses of 200, 400, 600 and 800r. Impressions of normal spleens were taken on the 11th, 12th, 14th and 16th days of incubation, and those of irradiated ones on the same days except the 11th.

The impressions were stained by the method of May-Grunwald-Giemsa, and we followed statistically the evolution of the spleen cells for each dose and for each latency, using 10 embryos in each case. Counting the cells in comparable microscopic fields, we established the percentage of maximal, medium and minimal numbers of the various categories of irradiated cells as compared to the normal ones of the same age.

Hemopoiesis in the chick embryo spleen is strongly perturbed already by doses of 200 r. The stem cells show lesions with 400 r. The lymphocytic strain is the most sensitive; its percentage diminishes beginning with 200 r. The erythrocytic strain is affected only with a dose of 600 r.

However, even with 800 r the lesions are not definitive. Hemopoiesis resumes on the 16th day for the lymphocytic and granulocytic strains; in the latter hyperbasophiles had been formed. The erythrocytic strain remains somewhat deficient.

In all cases there occur 3 phases in the cell divisions: (a) atypic mitoses, with double metaphases or anaphases, or double telophases with chromatic bridges between the filial nuclei; (b) a phase of mitotic arrest, which explains the cellular rarefaction; (c) a phase of resumption, apparently of normal mitoses, towards the 5th day following the irradiation.

188

H. M. MEISNER, University of Alberta. The effect of propylthiouracil on oxygen consumption and radioiodine metabolism of the frog, *Rana pipiens*, in winter. (Introduced by C. P. Hickman, Jr.) (15 min.)

This study was undertaken to evaluate the physiological importance of the thyroid gland in the adult frog, *Rana pipiens*, in winter. Male frogs, weighing between 40-55 g, were kept in running water and fed liver once a week. The thyroid gland was chemically inhibited by oral administration of 1.5 mg of propylthiouracil 3 times a week. Oxygen consumption was measured weekly from December 21 to February 23 with volumetric respirometers at a temperature of 16 $\pm$ 0.1°C.

There were never any significant differences between the metabolic rates of the propylthiouracil-inhibited frogs and the controls over the entire experimental period. Propylthiouracil effectively blocked the thyroid radioiodine uptake and blood conversion ratio, but after 9 weeks, thyroid iodine stores were only reduced by one-half. This low secretion rate together with the fact that propylthiouracil did not alter metabolic rate suggests that the frog thyroid is of no calorigenic importance during the winter.

The seasonal thyroid activity pattern was found to be characterized by a regression in the iodine-concentrating capacity of the thyroid gland and blood from August to December, and reactivation towards the end of February.

The appearance of monoiodotyrosine and diiodotyrosine in the circulation of *Rana pipiens* indicates that thyroidal deiodinase is unable to reduce iodotyrosines to iodide. It is postulated that there is an association between the presence of iodotyrosines in the circulation and the reduced thyroid function of poikilotherms.

105

DEFOREST MELLON, Jr. and DAVID R. EVANS, Johns Hopkins University. Electrophysiological evidence that water stimulates a fourth sensory cell in the blowfly taste receptor. (15 min.)

Associated with the labellar chemosensory hairs of the blowfly, *Phormia regina*, previous histological and electrophysiological studies have revealed three sensory cells for which adequate stimuli are NaCl, fructose, and bending. Recent physiological studies of thirst (Evans, Nature, in press; Dethier and Evans, in prep.) required that water stimulate an additional chemosensory cell. Accordingly, we sought and obtained electrophysiological evidence for the existence of a fourth sensory cell, an adequate stimulus for which is water. Recording from the side of a hair (Morita, J.C.C.P., 1959), impulses from the four cells were distinguished on the basis of spike height and shape, electrical summations and refractory period. Pure water is the most effective stimulus that we have assayed for the water re-

ceptor, and only that cell is activated by water. Aqueous solutions of non-electrolytes appear to depress the activity of the water cell as a linear function of the log. of the osmotic pressure, the frequency of firing decreasing to less than half of the maximum at five molal sucrose. Solutions of inorganic electrolytes also inhibit the water response, but this effect appears more specific. Aqueous NaCl solutions inhibit the water response first at about 0.1 molal and completely by 0.3 molal. $CaCl_2$ solutions abolish the water response at 0.01 molal. The inhibition of the water response is unrelated to spike activity in other fibers. Other investigators have observed the water spikes in response to dilute salt solutions but attributed it to the sugar fiber. (Aided by grant E-2358 and a predoctoral fellowship GF-9124-C2 (to DeF. M.) from the U.S.P.H.S.)

164

ROBERT J. MERKLIN, Jefferson Medical College. Ovarian, uterine and adrenal lipids of lactating mice. (15 min.)

While the corpora lutea of pregnancy (CLP) and lactation (CLL) each display a moderate amount of neutral fat in early stages of lactation, they differ greatly in phospholipid content. The former gives a strong reaction for phospholipids and the latter are essentially negative. By the twelfth day of lactation a difference in neutral fat content is also seen; the CLP exhibit a very light neutral fat reaction while the CLL give a strong reaction. Cholesterol content which was low or negative in all corpora lutea earlier, has a moderate to heavy distribution in the CLP at 12 days. Unsaturated lipids are seen only in the CLP of the ninth day of lactation. A strong reaction for glycoprotein is always seen in the zona pellucida and follicular fluid but is essentially absent from the rest of the ovary during lactation. The adrenal gland has a zone of degenerating cell clumps at the corticomedullary junction at the sixth day of lactation and these clumps are positive for neutral fat, cholesterol and glycoprotein. At the ninth day these clumps have disappeared and a firm boundary is seen between cortex and medulla. At this time the cortex has a uniform and moderate distribution of neutral fat, cholesterol and unsaturated lipid. At the twelfth day neutral fat and phospholipid in moderate quantities are seen in the zona fasciculata. The uterus is characterized by the presence of a metrial gland and a distribution of neutral fat, cholesterol and glycoprotein in this structure and in certain stromal elements.

238

J. F. METCALF and J. R. FISHER, Florida State University. Glutaminase activity in the brain of developing chick embryos.

Enzyme activities are being studied in embryos in order to describe developmental changes, correlate these changes with other events and to determine the mechanisms which control activity. Glutaminase activity has been measured in the brain of developing chick embryos and correlated with electrical activity. Evidence has been obtained that brain contains a glutaminase inhibitor and it appears possible that this agent is involved in controlling activity.

Electrical activity is essentially absent in the cerebrum, optic lobes and cerebellum of chick embryos on the 11th day of development and increases strikingly between the 13th day and hatching (Peters *et al.* Physiol. Zool. 33, 225). The amplitude is highest in the cerebrum and lowest in the cerebellum. These developmental changes and differences in amplitude were correlated with glutamotransferase activity. Sisken *et al.* (J. Biol. Chem. 236, 503) have shown a progressive increase in the concentration of γ-aminobutyric acid and in the activities of glutamic decarboxylase and γ-aminobutyric acid-α-ketoglutarate transaminase in the optic lobes of chick embryos during development.

Glutaminase activities of tissue homogenates were measured at an optimal pH (8.4), phosphate concentration (0.2 M) and substrate concentration (0.05 M). Ammonia production was followed colorimetrically. Activities increased from 14 μgs glutamine hydrolysed per hour per mg tissue in all three tissues on the 11th day to 96 for cerebrum, 54 for optic lobes and 23 for cerebellum on the day of hatching. These results correlate very closely with changes in electrical activity. (Supported by grant RG-4667 from the U.S.P.H.S.)

27

AKIO MIYAKE, Indiana University and Osaka City University, Osaka, Japan. Artificial induction of conjugation by chemical agents in *Paramecium* of the "*aurelia* group" and some of its applications to genetic work. (Introduced by R. V. Dippell) (15 min.)

Miyake (1956, '58, '60) has shown that chemical agents can regularly induce conjugation between animals of the same mating type in *P. caudatum*, *P. aurelia* and *P. multimicronucleatum*. The agents effective on the two latter species, K+acriflavine (or proflavine)+acetamide in Ca-poor media, are also effective on *P. jenningsi*. Those effective on *P. caudatum*, K+acetamide in Ca-poor media, are also effective on *P. traunsteineri*. All 5 of these species belong to the "*aurelia* group"; chemical induction of conjugation has thus far failed in paramecia of the "*bursaria* group" (*P. bursaria*, *P. trichium*, *P. polycaryum*, *P. calkinsi*). The chemical treatments have induced conjugation within several stocks that are pure for one mating type: in *P. aurelia*, stock P of syngen 1 (pure for mating type I) and stock 38 of syngen 7 (pure for mating type XIII); in *P. multimicronucleatum*, stock 30 of syngen 2 (pure for mating type IV). However, the same method failed to induce conjugation in the mutant stock CM of *P. aurelia* which can undergo the mating reaction but cannot form tight pairs. After induced selfing of stock 30, segregation of mating types III and IV seems to be caryondial, as earlier shown by Barnett in breeding experiments. Unions in which cross-fertilization is prevented by inappropriate pairing also occur in chemically-induced conjugation. In them, each conjugant probably undergoes self-fertilization and would be expected to become completely homozygous if sister gamete nuclei fused. Such unions were obtained in *P. multimicronucleatum*, syngen 2, in which natural autogamy is not known and some exconjugant

clones were viable; but proof of their homozygosity is still lacking. (Supported by a U.S.P.H.S. Traineeship, 2G-82 (CISI), and by a grant, AT (11-1)-235, to T. M. Sonneborn from the A.E.C.)

80

VALERIO MONESI, Biology Division, Oak Ridge National Laboratory and Biology Division of the National Committee for Nuclear Energy, Research Group for Embryology, Frascati (Roma), Italy. Study of DNA synthesis and the mitotic cycle in spermatogonia of the mouse, using tritium-labeled thymidine. (Introduced by E. F. Oakberg) (15 min.)

C3H♂ × 101♀ F_1 hybrid male mice were injected intraperitoneally with 15 μc of H^3-thymidine per mouse. Using autoradiography, the time course of the labeling in spermatogonia was studied through two successive mitotic cycles.

Four generations of type A, one of intermediate and one of type B spermatogonia occur in one cycle of the seminiferous epithelium. The average generation time is about the same in all six generations, or about 27 to 30 hr. The average pre-DNA synthetic time, including the mitotic stages from metaphase through telophase and the portion of interphase preceding DNA synthesis is also uniform, or 8½ to 10½ hr. A remarkable difference exists, however, in the duration of DNA synthesis and of the post-DNA synthetic period. DNA synthesis in spermatogonia occurs throughout the second half or the last ¾ of interphase, according to the type of spermatogonium. The average DNA synthetic time is very long with high variability in type B (14 to 15 hr) and in intermediate (12 to 13 hr) spermatogonia and gradually shorter with lower variability from the fourth (12 hr) to the first type A generation (7 hr). Conversely, the average post-DNA synthetic time, including prophase and part of metaphase, is short and very constant in type B (4½ hr) and intermediate (6 hr) spermatogonia and gradually longer and more variable from the fourth (8 hr) to the first type A generation (14 hr).

The meiotic synthesis of DNA occurs in primary spermatocytes throughout the second half of interphase. Its average duration is 14½ hr. No further synthesis of DNA takes place in later stages of spermatogenesis.

39

A. ULRIC MOORE and HELEN BLAUVELT, Cornell University. Behavior during a conditioned reflex training period used as a measure of early trauma in lamb and kid. (15 min.)

The pattern and amount of movement in an open room is recorded during a 40-minute period of two-minute-spaced, always-reinforced, ten-second signals and a comparison is made between the behavior of 20 experimental animals and a like number of controls during their first four periods of conditioning.

The experimental group (11 lambs and 9 kids) were in various ways "stepchildren" ("cinderellas") or were step-siblings, some even having been raised by a mother of the other species (kids by sheep and lambs by goats). On the basis of several other criteria it is assumed that the trauma produced is of a very early and profound nature, involving a subversion of the source of food and comfort. The only source of food (and hence of life) becomes also a source of fear. The resultant behavior is striking in the young experimental animals and can easily be differentiated even after two years by the conditioned reflex method.

The change in the pattern and amount of movement during a conditioned reflex training period appears to depend far more upon the degree of normality or abnormality in the relation of the infant to its mother figure than upon any subsequent experiences the young animal may have. Wide variations in such experiences and in individual differences such as sex, size, dominance, species, age at testing etc. for both controls and "cinderellas" make for little change in this conditioning parameter, whereas seemingly slight variations in mother-neonate relations alter the parameter drastically. (Supported in part by grants from the Josiah Macy, Jr. Foundation and the Ford Foundation.)

9

ROBERT E. MOORE, University of Texas. Olfactory discrimination by individuals of *Peromyscus maniculatus* and *Peromyscus polionotus*. (Introduced by W. F. Blair) (15 min.)

There is experimental evidence for sexual isolation between *Peromyscus maniculatus* and *Peromyscus polionotus* (Blair and Howard, 1944), and experiments were devised to determine if olfaction plays a role in species recognition and sexual isolation. Single individuals of each species were given a choice of three similar compartments which were connected by tunnels. An individual of the same species as the one being tested for olfactory discrimination had previously occupied one compartment, and an individual of the other species had previously occupied a second compartment. The movements of the individual being tested were recorded by microswitches and electromagnet arms on a slow-moving roll of paper, and the preference of compartments was correlated with the previous occupancy. (Supported by fellowship 20200 from the N.S.F.)

67

LOUIS G. MORIBER, LAWRENCE PRUTKIN and MARK E. PARKS, Brooklyn College. The nature of differential staining with acid dyes. (15 min.)

Triple staining techniques generally include acid dyes, basic dyes, and mordants. We have studied a wide variety of groups of acid dyes without mordants, utilizing either a series of staining baths each composed of one dye in 2% acetic acid or one bath containing a mixture of acid dyes in 2% acetic acid. Different dye concentrations and immersion times were required depending on fixation. Naphthol yellow S, acid fuchsin, and methyl blue after Elftman's fixative (Stain Tech. 32: 25, 1957) produced particularly excellent results. For example: thyroid colloid is yellow; collagenous fibers are blue, but perichondrium is red; erythrocytes are yellow while plasma is red; muscle "A" bands are dark blue, "I" bands pale yellow; adenohypophyseal cell cytoplasms and nuclei are stained differentially.

At pH2 staining with anionic dyes is attributed to salt linkages with protein basic groups. Acetylation or deamination pretreatment reduced stainability generally, virtually eliminating naphthol yellow S. Hot TCA pretreatment intensified staining even of nucleic acid-free components, acid fuchsin staining most avidly.

Among those factors purported to play a role in such differential staining are the numbers and kinds of basic groups, texture of the protein and the degree of polymerization of the dye molecules, the pK's of the dyes, and dye molecular weights. From studies conducted to date, particularly differential binding of anionic dyes from solutions of 2 or more dyes, the authors favor the view that the most important factors are the size of the dye molecule and the development of secondary chemical bonding (van der Waals) with protein side chains.

239

JOHN B. MORRILL, Wesleyan University. Development of *Lymnaea stagnalis* eggs with giant polar bodies.

Encapsuled eggs were centrifuged at 4800 rpm for 10 minutes 20 to 10 minutes before second polar body (pb) formation. That the first pb was over the oil cap at the centripetal end of the elongated eggs indicated the eggs tended to orient with their animal-vegetal axis parallel to the centrifugation axis. Following centrifugation a pb 30 to 115 μ in diameter appeared beneath the 1st pb. Of 32 eggs with a 2nd pb 30 to 60 μ in diameter 75% developed into normal snails and 25% died in the cleavage or gastrula stages. Of 35 eggs with a 2nd pb 65 to 85 μ in diameter 37% developed into normal snails; 9% into snails with normal heads but abnormal shell glands, feet and masses of larval liver cells; 14% into veligers with paired cephalic plates but abnormal shell glands, feet and masses of larval liver cells; and 40% died in the cleavage or gastrula stages. Of 29 eggs with a 2nd pb 90 to 115 μ in diameter 3% developed into normal snails (2nd pb 90 μ in diam.), 3% into veligers; and 94% died in the cleavage or gastrula stages. These results demonstrate that stratified, uncleaved eggs in which the oil cap and as much as 25 to 30% (2nd pb 85-90 μ in diam.) of the egg's mass and cortex including the animal pole cortex is removed *via* a giant polar body may develop normally.

The data suggest that if an animal gradient field in the cortex controls the normal pattern of head organs (Raven & Beenakkers, '55), the field is not present before 2nd pb formation. Of if present, the field regulates following removal of the animal pole cortex; and/or the center of the field is not localized at the animal pole. (This study was made during the tenure of a N.S.F. postdoctoral fellowship while a member of the 3rd International Teamwork on Embryology at the Hubrecht Laboratory, Utrecht, Netherlands.)

240

JAMES E. MORROW, University of Alaska. Zoogeography of *Chauliodus* (Pisces, Clupeiformes).

Systematic revision and new locality records suggested former concepts of zoogeography of this genus also need revision. In the North Atlantic, as Haffner (Syst. Zool., 1: 112-133, 1952) showed, *C. sloani* and *C. danae* have different oxygen requirements. In the south Atlantic, *C. sloani* and *C. schmidti* have similar oxygen requirements during daylight, but in darkness latter species is found chiefly above 80% and below 30% saturation. Latter species also inhabits central part of South Atlantic Central water mass, but *C. sloani* occupies water of greater or lesser salinity.

C. macouni is confined to Pacific Subarctic water and to transition zone southward along North American coast, characterized by low salinity and temperature. *C. barbatus* is found only in eastern part of Pacific Equatorial water. *C. sloani* occurs throughout Equatorial and Central water masses of Pacific and Indian oceans. There is no physicochemical basis for assuming isolation of sub-specific forms.

Small light organs in row between serial organs become progressively reduced from east to west in Pacific and Indian oceans and into Atlantic, in *C. sloani*, and also from tropical to temperate regions. Length of chin barbel is reduced in western areas as compared with eastern Pacific. This and other evidence suggests that *C. barbatus* represents a separate line of evolution within the genus and is a relict form. All other species are derived from the basic *sloani* stock, which most probably arose not in the Indo-Australian region but in the eastern Pacific.

24

JO ANNE MUELLER, Indiana University. Further studies on the nature of the "co-factor" required for kappa infection in *Paramecium aurelia*, syngen 4. (Introduced by D. G. Frey) (15 min.)

Sensitive animals placed in a high concentration of disrupted killer animals become infected and, under proper conditions, develop into killers (Sonneborn, 1950). If the brei of disrupted killers is centrifuged at 25,000 $\times$ g for 5 minutes, all kappa particles are packed in the sediment. An infection test set separately with sediment and supernatant yields little or no infection, but if the fractions are recombined, infection is readily obtained (Tallan, 1959). The present experiments investigate the nature of the "co-factor" in the supernatant. The assay to detect the presence or absence of "co-factor" involves exposing 200 sensitive detectors in 0.2cc of fluid to infective mixtures. A dilution series of particles + "co-factor" is made and compared to a dilution of particles only, keeping the "co-factor" constant. If "co-factor" is in excess, no dilution effect is obtained in the infection tests throughout the range employed; but if "co-factor" is diluted, then the percent of successful infective mixtures follows the dilution factor. The supernatant from a brei of sensitives with the K- or kk genome, as well as that from killers, both contain "co-factor." $CaCl_2$ in a sublethal concentration serves as an efficient "co-factor." A dilution of the salt results in a reduction of infectivity. Although preliminary work suggests certain other salts may be effective as "co-factor," substances such as Carbowax 4000, Lysozyme, NaH_2PO_4 and L-tryptophan, in the concentrations investigated, are not active. (Supported by American Cancer grant E 81 B to Sonneborn.)

187

PHILIP F. MULVEY, Jr. and D. WARD SLINGERLAND, VA Hospital, Boston. Stimulation of the hexose monophosphate shunt in rat thyroid slices by small amounts of propylthiouracil. (15 min.)

Previous observations that small amounts of propylthiouracil increased the uptake of radioiodine and the consumption of oxygen by rat thyroid slices led to a study of the effect of propylthiouracil on the metabolism of C^{14} labeled glucose. A high concentration of propylthiouracil (10^{-3}M) in the medium diminished the production of $C^{14}O_2$ by rat thyroid slices at one hour, from both glucose-1-C^{14} and glucose-6-C^{14}. However, with small doses of propylthiouracil (10^{-5}M), the production of $C^{14}O_2$ from glucose-6-C^{14} was markedly stimulated (15 fold; $P = 0.025$) whereas the production of $C^{14}O_2$ from glucose-1-C^{14} was still inhibited ($P < 0.005$).

Thyrotropin stimulated the oxidation of glucose-1-C^{14} to $C^{14}O_2$ and, to a lesser extent, the production of $C^{14}O_2$ derived from glucose-6-C^{14}.

In summary, small amounts of propylthiouracil (10^{-5}M) appear to stimulate not only the uptake of radioiodine and oxygen consumption by rat thyroid slices, but also the Embden-Myerhof glycolytic pathway, which TSH stimulates to a lesser extent, suggesting an independent stimulatory action of this antithyroid compound.

151

DARREL MURRAY and EARL SEGAL, Purdue University and Rice University. The effects of temperature and light on locomotor activity of the American cockroach, *Periplaneta americana*. (15 min.)

Cockroaches, *Periplaneta americana*, were maintained at saturated humidity for one week under the following conditions: (1) constant temperatures of 10°, 20° and 30°C and in constant light, constant dark and alternating light-dark (LD 11:13) for each temperature; (2) alternating temperatures of 10°-30°C (12 hours at each temperature) and 20°-30°C and in constant light and constant dark. Total locomotor activity during a 24-hour period at 30°C in constant light was recorded by measuring increase in light transmission through smoked glass plates over which animals crawled. All recordings were initiated at the same time of day (10 A.M.).

Animals maintained at 30°C in constant light and constant dark were less active than 20°C animals and more active than 10°C animals. In alternating light-dark, 30°C animals were more active than either 20° or 10°C animals. Light conditions had no significant effect on activity of 10° and 20°C animals. At alternating temperatures of 10°-30°C, animals kept in constant dark were more active when maintained in the cold (10°C) for the 12-hour period prior to recording than animals maintained in the warm (30°C); similar results were obtained for animals maintained at 20°-30°C. Differences were not significant between groups maintained at alternating temperatures in constant light.

The data indicate that light conditions have no significant effect on the activity of *Periplaneta americana* at constant temperatures of 10°C and 20°C but the effect is significant when animals are maintained at 30°C. It was concluded that immediate thermal history is important in determining activity under conditions of alternating temperature.

241

X. J. MUSACCHIA and MELCHIOR ISTVAN CHLADEK, St. Louis University. Investigations of the cloacal bladders in turtles.

The functional role of cloacal bladders in Chelonia has seldom been explored experimentally. Aspects of two reputed functional roles, (1) underwater respiration and (2) water storage, were studied in *Chrysemys picta* and *Terrapene carolina*.

In order to utilize cloacal bladders for respiration, water must be taken in and ejected from the bladders. *C. picta* were (muzzled and unmuzzled) submerged in water containing carmine and neutral red. The dyes confirmed entrance of environmental fluid; fluid volume of cloacal bladders was also recorded. Some summarized results are: 20 specimens muzzled and unmuzzled, (2°C, 15 days) didn't take in water; of 8 turtles (5°C, 50 days), 5 muzzled and 2 unmuzzled specimens took in water, 1 unmuzzled did not; of 10 muzzled turtles (10-12.5°C, 6 days) 7 took in water, 3 did not; of 22 turtles (26°C, 2 to 22 hours), 17 muzzled and 5 unmuzzled, 15 and 4, respectively, took in water. The weight of evidence is that in the successively higher environmental temperatures, more turtles reacted by taking water into cloacal bladders. These findings establish that water can be taken into cloacal bladders and indirectly corroborate a presumed respiratory function.

T. carolina, a "terrestrial" turtle, was used experimentally to evaluate "water storage" function of cloacal bladders. Twenty-nine specimens were fasted under arid conditions (5 to 15 days, 32°C), then placed in water and permitted to drink *ad libitum*. Water intake volume was measured and, from 3 to 24 days later, animals were autopsied. In each animal the cloacal bladders were empty. Cloacal bladders in *T. carolina* are small; however, some submersion experiments resulted in filled, distended cloacal bladders. It was concluded that in *T. carolina* cloacal bladders do not function in water storage.

152

JOHN A. MUTCHMOR and A. GLENN RICHARDS, University of Minnesota. Low temperature tolerance of insects in relation to the influence of temperature on muscle apyrase activity. (15 min.)

The influence of temperature on the rate of apyrase activity was examined in arthropod species that differ in chill-coma and habitat temperatures. At given low temperatures the rates and temperature coefficients of apyrase activity were generally higher in animals with the lower chill-coma temperatures. However, at temperatures 3-4°C above the respective chill-coma temperatures, the rates were nearly the same in each species (0.4 ± 0.1 μg P/mg muscle/min). The relationship between chill-coma temperature and temperature coefficient was linear; in the insects 1°C difference in coma temperature was equivalent to a difference of about 2,400 cal in μ-value.

Apyrase rate maxima occurred at lower temperatures in species having lower chill-coma temperatures, and the maxima were greater in species that might be regarded as normally most active. Cold adaptation in the American cockroach resulted in a higher apyrase rate but had little or no effect on the temperature coefficient of apyrase activity.

It was concluded that (1) higher apyrase rates, (2) higher temperature coefficients in species with lower chill-coma temperatures and (3) the occurrence of a common rate of apyrase activity at a temperature several degrees above the respective chill-coma temperatures, might well constitute important mechanisms involved in the adaptive compensation permitting maintenance of activity at low temperatures. The data are consistent with the suggestion that perhaps a minimum rate of hydrolysis of ATP is necessary to permit muscular contraction.

242

R. NAGABHUSHANAM, Andhra University. Response of marine borers to chemically treated timbers.

The present report deals with the results of experiments carried out in Visakhapatnam Harbour, India, during the year 1956 on the response of marine wood-boring mollusks to various wood preservatives. Two types of timbers, *Pinus longifolia* and *Myristica fragrans*, were selected for chemical treatment. Inspections were made once a month, after taking the specimens from water and removing all the foulers that had collected on their surfaces. In all tests the controls were run simultaneously and they were completely destroyed in two months.

Among the 12 preservatives used in these experiments, the most effective are the Ascu (concentrations ranging from 1.0 to 2.0 lbs dry salt absorbed per cu ft), Creosant (15 lbs solution absorbed per cu ft), Creosote (21.6 lbs solution absorbed per cu ft) and Copper abietate (0.163 to 0.203 lbs dry salt absorbed per cu ft). The response of the pholad, *Martesia striata*, and shipworms to certain compounds was quite different. Creosant and Creosoted wood inhibited the action of shipworms during the whole period of investigation, whereas *Martesia* often seemed to thrive in the timbers treated with these products.

243

R. NAGABHUSHANAM, Andhra University. Ecological factors related to the distribution of *Bankia campanellata* in Visakhapatnam Harbour.

The most abundant species of *Bankia* in Visakhapatnam Harbour, India, is *Bankia campanellata*. The local distribution of this species was determined by differences in its relation to ecological factors for the period 1955-56. *Bankia* made its appearance in test panels in August, 1955 and continued to be present till the end of February, 1956. In summer between March and July when the temperature was comparatively high (30.9°C) not even a single *Bankia* was noticed in the monthly test panels. It was quite probable that this form might not be breeding during these months. In August, 1956 when the temperature started declining the animals began to appear and gradually increased in number and reached their maximum in December-January period when the water temperature was lowest for the year (26.5°C).

The available evidence on vertical distribution showed that the largest portion of the total number of *Bankia* was usually found in the lower half of the vertical column. This species was found in the harbour at salinities between 21 and 34%. The magnitude of strike decreased with decreasing salinity.

77

DONALD J. NASH and JOHN W. GOWEN, Pennsylvania State University and Iowa State University. Reproductive performance of mice exposed to embryonic X-irradiation. (15 min.)

Mouse embryos of nine different genotypes (consisting of progeny of three inbred strains, BALB/Gw, K, and S, and the hybrids derived from them) were exposed *in utero* to single whole-body X-ray (250 pkv, 30 ma with 0.25 mm Cu and 1 mm Al filtration) dosages from 20 to 320 roentgens on 6½, 10½, 14½ or 17½ days gestation as timed from the appearance of a vaginal plug. At two and a half months of age two males and two females of each genotype were tested for lifetime reproductive performance within each dose-embryological age combination. Each treated mouse was caged separately with a nonirradiated Z strain mate for the lifetime of the treated mouse. Results of matings one year from the beginning of the experiment are reported here for the overall mouse population. Fertility as judged by the mean number of progeny born in the one year period was found to be dependent both on the level of radiation and embryological age at the time of exposure. Embryos that had received 80r, for example, at 10½ days had only 84% as many progeny as controls whereas the fertility of mice irradiated with up to 160r at 6½ days wasn't affected. Embryological ages in order of increasing sensitivity were 6½, 17½, 14½ and 10½ days. These results indicate that the response of the fetal reproductive system to radiation doses varies significantly with the age at which the radiation exposure was received. (This work has received assistance from Contract AT (11-1) 107 from the A.E.C. Journal Paper No. J-4116 of the Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa. Project No. 1180, 1187.)

37

T. L. NASH, J. M. WARREN and M. W. SCHEIN, Pennsylvania State University. Effects of time and interpolated problems upon retention in chickens. (15 min.)

Six White Leghorn hens, previously trained on simple discrimination learning, were used to test retention and retroactive effects in nonspatial discrimination learning. Stimulus objects were pairs of wooden, metal and plastic figures varied in multiple visual dimensions. Cue pairs were presented daily using a mechanically manipulated apparatus, employing food as a reward. Subjects were permitted immediate correction. Median savings scores of 80-95% were obtained over individual sets of 1, 4 and 10 interpolated problems. Three sequences

of 18 interpolated problems yielded 8-49% savings. A definite increase in trials and errors to criterion resulted from reversals of original discrimination problems after 18 interpolated sets. Time interval over the testing period appeared to have relatively little influence upon retention. It is concluded that the subjects demonstrated a hitherto unsuspected ability to remember for at least 19 days the solution to a problem despite the intervening imposition of up to 18 other problems.

117

CHARLES NORMAN, C. E. JOHNSON, I. D. PORTERFIELD, E. GOLDBERG and R. S. DUNBAR, Jr., West Virginia University. Prolonged maintenance of mammalian sperm in a chemically defined medium at room temperatures. (15 min.)

A chemically defined medium, Norman and Johnson's solution (N-J), has been developed for prolonged maintenance of physiologically active human and bovine sperm at room temperatures (21°-27°C). The medium is composed of 2.16 g % sodium citrate dihydrate, 500 mg % glucose, 482 mg % calcium nitrate tetrahydrate, 45 mg % magnesium sulfate heptahydrate, 34 mg % sodium sulfate, 14 mg % potassium nitrate, 11 mg % potassium chloride, 3 mg % monosodium phosphate, 300 mg % sulfanilamide, 68 mg % dihydrostreptomycin sulfate, 31 mg % penicillin G (Na), 150 units sterile aqueous solution catalase/ml, and 4 units mycostatin/ml. For use in artificial breeding programs, inclusion of 1.0% egg yolk is usually necessary to protect bovine sperm against cold shock.

The well-defined nature of the N-J medium enlarges the scope of possible research on the effects of heredity and environment on the *in vitro* survival of post-mitotic mammalian cells. (Supported by grant RG-6339 from the N.I.H. and by a grant from the N.A.A.B.)

244

CHARLES NORMAN, J. L. McCOY, I. D. PORTERFIELD and H. A. WILSON, West Virginia University. Elimination of *Vibrio fetus* from bovine sperm stored at low temperatures.

The effect of a pre-freezing incubation period on *Vibrio fetus* inoculated sperm subjected to dihydrostreptomycin, neomycin and chloromycetin was observed. Each antibiotic was added singly to aliquots of coconut milk extender containing *Vibrio fetus* (1.44×10^8 organisms/ml) and 4.4×10^7 sperm/ml. In addition, control aliquots without antibiotics and aliquots with antibiotics but no spermatozoa were allowed to incubate for 2 and 6 hours at 30°C and for 24 hours at room temperature (25°C ± 2°). After incubation, samples were frozen to –79°C. After a week of storage at –79°C, (1) the extent to which *Vibrio fetus* could be recovered and (2) sperm motility were determined for each sample. The results of this study indicated dihydrostreptomycin was bactericidal at a 680 μg/ml level after all incubation periods. Neomycin was bactericidal at a 250 μg/ml level after 24 hours incubation at room temperature. A 250 μg/ml level of chloromycetin showed no bactericidal effect. The pre-freezing incubation periods produced no measurable effect on the viability or motility of the sperm cells. These data indicate that an appropriate pre-freezing incubation period can eliminate *Vibrio fetus* from bovine sperm suspensions treated with dihydrostreptomycin or neomycin without harming the spermatozoa. (Supported by grant E-2497 from the U.S.P.H.S.)

125

DAVID A. NUNNALLY, Vanderbilt University. Development of succinic dehydrogenase in the duodenum of the chick. (Introduced by Florence Moog) (15 min.)

Succinic dehydrogenase (SDH) was determined by colorimetric estimation of formazan produced when neotetrazolium chloride was incubated with succinate and homogenates of whole duodenal wall of chick embryos from 15 days to hatching, and of chicks up to 29 days of age. The enzyme activity (expressed as μg formazan produced/mg protein) is at a uniformly low level until the time of hatching, when it begins a rise to a peak at five days; the maximal level is about 8 times the embryonic level. Activity then drops to about 2/3 the 5-day level, where it remains throughout the period under study. Histochemical tests with neotetrazolium indicate that practically all the SDH activity is in the epithelium. When 1 mg of cortisone acetate was injected into the eggs at 15 days, there was a slight but significant ($P < .05$) increase in SDH by 17 days, maintained at 18 and 19 days. At hatching, there was no difference between experimentals and uninjected controls. When 3.8 mg of thiourea was injected at 11 days, SDH activity at 20 days was depressed not only with respect to the controls, but also when compared with experimentals at 18 days. Thiourea-treated embryos never hatched by 26 days of incubation, and there was never any increase in intestinal SDH during this time. This is in contrast to alkaline phosphatase, which begins to rise belatedly in thiourea-treated chicks, but does not attain normal levels. Injection of succinate into incubating eggs failed to elevate SDH activity. (Supported by a N.S.F. Pre-Doctoral Fellowship, 1959-60, and by U.S.P.H.S. grant RG-3937 to Dr. F. Moog, Washington University.)

50

ROBERT E. OGREN, Dickinson College. A contribution to the biology of the land planarian, *Microplana atrocyaneus* (Walton, 1912). (10 min.)

This large black or purple land planarian, usually considered rare, was found beneath logs in two new localities: Carlisle, Pa., and Jamestown, N. Y. The largest specimen, 25-30 mm long when extended, was the most mature specimen available for study. Condition of the mature reproductive apparatus was observed. In the mature specimen a long, twisted, ciliated genito-intestinal canal opened into the intestine. The eosinophilous glands opened into the vagina before the origin of the genito intestinal canal. Numerous testes were present and the penis papilla was well developed.

Mature specimens were black; immature ones about 10 mm long, a mottled purple. During movement the first 2 mm of the elongate body were

held above the substrate to form a short rounded proboscidiform anterior portion. This area contained the small eyes and was waved from side to side. Locomotion was performed by a modified form of myopodal progression. Response to bright light was very slow. The rate of movement was slower and the degree of response to touch was poorer than that of the smaller, awl-shaped land planarian, *Rhynchodemus sylvaticus*. (Partial support from N.S.F. grant G12905.)

245

MARK E. PARKS and LOUIS G. MORIBER, Brooklyn College. A freezing-substitution method permitting the cytochemical demonstration of lipids.

The freezing-substitution method usually employs alcohol or acetone as the substitution medium. Ethylene glycol, or similar media which do not dissolve lipids, freeze or are extremely viscous below –35 to –40°C, the temperatures above which ice crystal growth seriously damages the otherwise well preserved microscopic structure. Therefore, our attention was directed toward mixtures of ethylene glycol, propylene glycol and glycerol, utilizing thereby the principle of mutual depression of freezing points. Other substitution media incorporating, in addition to the glycols, a mixture of alcohols were formulated further to reduce viscosity and freezing points.

Lipid solubility in these various media was investigated first by immersing narrow bore tubes filled with a dyed oil at room temperature for extended periods of time. It was observed that there was no detectable evidence of solubility in the glycol mixtures even at alcohol concentrations of 30% or higher.

Tissue blocks were then frozen in the usual manner in isopentane cooled with liquid nitrogen and transferred to various substitution media maintained at –40 to –50°C for periods of 5-7 days. After reaching room temperature, the dehydrated tissues were passed through several changes of molten Carbowax embedding mixtures and cast into blocks. Sections were floated on ethylene glycol and blotted carefully. Slides were dried in an evacuated dehydration chamber and then were stained with Sudan dyes in propylene glycol. Counterstaining with carmalum or hematoxylin proved satisfactory. Other routine staining techniques are compatible with this method. Enzyme activity has not been studied as yet.

8

ROGER S. PAYNE, Cornell University. The acoustical localization of prey by the Barn Owl *(Tyto alba)*. (15 min.)

Barn owls, *(Tyto alba)* can locate prey in total darkness using hearing alone, with an accuracy of about one degree in the vertical and horizontal planes. Measurements of sound pressure in the region of the owl's eardrum made with a probe tube microphone, while moving a loudspeaker around the owl's head, reveal that for frequencies of sound above 9,000 cps the ear is highly directional. At such frequencies, regions of high and low sensitivity are directed along different paths for the two ears. These are correlated with the asymmetry of owl's ears. By recording cochlear microphonics similar variations are obtained. Experiments with owls trained to strike a concealed loudspeaker show they depend on frequencies above 9,000 cps. The behavior of barn owls flying at prey in complete darkness differs from such behavior in the light. The changes are associated with the ability of the owl to locate prey acoustically. (This research was supported in part by the American Museum of Natural History, Frank M. Chapman fund; the Quartermaster Corps, U. S. Army (DA19-129-QM-1428); and by O.N.R. Contract (#Nonr-3225 (00).)

90

LOUIS J. PIERRO, University of Connecticut. Teratogenic action of actinomycin D. (Introduced by Walter Landauer) (15 min.)

Polymorphic malformations involving the viscera, skeleton, eyes, nervous and cardiovascular systems have been induced in mammalian embryos by treating pregnant females with actinomycin D during a critical period (Tuchmann-Duplessis and Mercier-Parot 1958-60). The studies to be reported demonstrate that this antibiotic is also teratogenic in the embryonic chick.

Injection of actinomycin D into the yolk sac of chick embryos induces anomalous skeletal development. Extremely abnormal embryos lack varying portions of the trunk, have abnormally positioned, defective hind appendages, if they are present at all, and may occasionally lack forelimbs. All of these embryos die around the eighth day of incubation. Less affected embryos are rumpless, and may exhibit defects of the hindlimbs and/or abnormal curvature of the pelvis. The frequency of abnormal embryos and extent of abnormality are primarily conditioned by the developmental stage treated, over the range of concentrations tested. Peak frequencies and the greatest proportion of extremely abnormal embryos are obtained following treatment during the middle third of the second day.

Presumably, actinomycin D selectively interferes with the development of the axial skeleton in the chick, limb and pelvic anomalies arising secondarily. The mechanism of action of this substance is obscure at present. Supplementation with sodium pyruvate, effective in protecting against certain types of chemically-induced rumplessness (Landauer and Rhodes 1952), is ineffective. Other supplements (*e.g.* calcium pantothenate) rather unexpectedly significantly increase the frequency of rumplessness.

246

PAUL PIETSCH, Bowman Gray School of Medicine and Wake Forest College. Effects of X-rays on regenerating limb and tail blastemata.

Tails of 30 mm *Amblystoma larvae* were amputated just distal to the cloaca. Animals were exposed to a single dose of 3000 r total body X-radiation 1-10 days after amputation. Control limbs and tails amputated after radiation did not regenerate. Dose was lethal but not within period of observation. Tail blastemata continued to grow

after X-radiation and regenerate size was approximately doubled. Tail regenerates radiated 7-10 days after amputation continued to grow at about 70% of normal rate and for two weeks. Then, quite suddenly, involution set in and within 2 more weeks the entire regenerate was resorbed. Measured planometrically, total amount of tissue accumulated after radiation was in direct proportion to days of regeneration before exposure.

Animals with 10-day regenerating limb blastemata were exposed to 1500 r total body X-radiation. Contralateral limbs amputated after radiation did not regenerate. X-radiated 10-day regenerates continued to grow for about three weeks, tripling preexposure size and developing aberrant hands with two or three incomplete fingers. A month after exposure aberrant hands began to involute and within a few weeks almost all regenerate tissue was resorbed.

Development of regenerating limb and tail blastemata was not brought to an immediate halt upon exposure to X-rays. There were no exceptions in over one hundred cases. However, involution invariably claimed virtually all of the regenerate including the portion formed prior to exposure. The findings seem to indicate: (a) there are some newly produced substances in the blastema which are not destroyed directly by X-rays; (b) there is a cumulative reaction set into motion following radiation that eventually leads to breakdown of newly formed tissue. (Supported jointly by Grant RG-7491, U.S.P.H.S. and IN-48A-4, American Cancer Society.)

247

LAJOS PIKÓ, California Institute of Technology. *In vitro* observation of cleavage of dispermic rat eggs.

Earlier observations (Austin and Braden, Aust. J. Biol. Sci., 6, 674, 1953; Pikó and Bomsel-Helmreich, Nature, 186, 737, 1960) indicated that dispermic rat eggs cleave normally and result in triploid embryos. To examine further the cleavage behaviour of dispermic eggs attempts were made to culture rat eggs *in vitro* after examination by phase microscopy.

Fertilized eggs were collected from 22 female rats of the "Sherman" strain which had been mated towards the end of estrus (1-2 hours after the average time of ovulation). Six of the females were subjected to a moderate hyperthermia (body temperature raised to 40-41°C during about 2 hours after mating) in order to increase the frequency of polyspermy. In cleaved eggs the evidence of dispermy was based on the presence of two sperm tails within the egg cytoplasm.

A total of 128 eggs, 8 of them dispermic, in the 1-, 2- and 4-cell stage was cultured in a variety of media (mostly in a phosphate-buffered balanced salt solution containing 1 mg/ml glucose and 1% human serum albumin). Many eggs, including some dispermic ones, made one further regular division (1st, 2nd or 3rd cleavage) *in vitro*, particularly if put into culture when cleavage *in vivo* was approaching. None underwent more than one cleavage *in vitro*.

An additional 33 eggs at the 16- to 32-cell stage (recovered 4 days after mating) were cultured in the presence of 0.5 gamma/ml colchicine. The eggs were then treated by a hypotonic saline, fixed, stained by aceto-carmine and squashed. Sixteen eggs furnished analyzable mitoses. Of these, 15 were diploid ($2n = 42$); in a dispermic egg however (with the remnants of 2 sperm mid-pieces in the egg) the two countable metaphase groups were triploid.

These data accord with earlier *in vivo* observations that dispermic rat eggs cleave like monospermic eggs, but are triploid. (Supported by a grant from the Lalor Foundation.)

248

LAJOS PIKÓ, California Institute of Technology. A study on the possibility of dispermic androgenesis in X-irradiated rat eggs.

Androgenetic development in eggs previously exposed to X-rays is a well-known phenomenon in Amphibia and in some lower animals. Viable androgenetic offsprings have been produced so far in the wasp *Habrobracon* and in silkworm; in the latter diploidy is achieved through the fusion of two sperm nuclei.

For examination of the possibility of occurrence of this phenomenon in the rat, where dispermy is not infrequent, the effects of different levels of X-irradiation on fertilization and cleavage were followed. Unmated estrous females were irradiated at 600, 1200, 2500, 6300 and 13,400 r towards the end of estrus when they contained tubal eggs in metaphase II. At 2500 r, and higher, the anterior half of the body was shielded. The females were mated immediately after irradiation and sacrificed within 3 days.

A total of 195 eggs, coming from 19 females, was examined by phase microscopy in the living state, and after aceto-carmine staining. Sperm penetration was normal although with the higher doses the proportion of fertilized eggs decreased (to 60% at 13,400 r). Emission of 2nd polar body took place and pronuclear development proceeded nearly normally in all groups; however, chromosomal bridges at second meiosis and female pronuclei of "teardrop" shape occurred. In many eggs, the female pronucleus failed to migrate to the middle of the egg, resulting in excentric spindles. Cleavage was delayed, especially after the higher doses. But the main anomaly was nuclear fragmentation at the first and later cleavages. Six dispermic eggs were observed. Three of these, obtained in the 6,300 and 13,400 r group, were in the 2-cell stage and their nuclei appeared more nearly normal than most of the monospermic eggs in these groups. Further studies of this subject are in progress. (Supported by a grant from the Lalor Foundation.)

16

C. A. PRIVITERA and H. J. MERSMANN, St. Louis University. Comparative oxidative phosphorylation by mitochondria from tissues of the turtle, *Chrysemys picta*. (15 min.)

Quantitative studies of energy metabolism in terms of mitochondrial oxidative phosphorylation have been grossly neglected among poikilotherms

with the exception of the work of Kanungo and Prosser, J. Cell. and Comp. Physiology, 1959.

Both liver and heart tissues of adult *Chrysemys picta* were homogenized in 0.25M sucrose in an ice jacketed Potter-Elvehjem homogenizer. The homogenate of each respective tissue was centrifuged according to modifications of the method of Schneider and Hogeboom ('50). Each discernible fraction obtained by centrifugation was assayed for its ability to mediate the process of oxidative phosphorylation using alpha-ketoglutaric acid as substrate (without a malonate block).

In the case of both the liver and the heart, only one particulate fraction resulting from centrifugation procedures proved to be capable of oxidative phosphorylation. All determinations were made in duplicate or triplicate with consideration for endogenous respiration.

The data obtained from the particulate liver suspension follow: μMP esterified/mg N/hr = 14.16 $\pm$ 3.00; μAO consumed = 7.20 $\pm$ 1.38/mg N/hr; P/O = 1.97. The data obtained from the particulate heart suspension follow: μMP esterified/mg N/hr = 123.57 $\pm$ 8.31; μAO consumed/mg N/hr = 51.27 $\pm$ 2.25; P/O = 2.44.

These data essentially indicate a quantitative base line of activity and strongly suggest that on a milligram of nitrogen basis, the heart is measurably more active in terms of energy metabolism than is the liver. An extended comparison between these data and data obtained from rat liver and heart will also be made. (Supported by grant from the Women's Advertising Club of St. Louis and A.M.A. grant 480A.)

88

WALTER C. QUEVEDO, Jr., JOHN M. CAIRNS and JEAN A. SMITH, Brown University and Roswell Park Memorial Institute. The effects of croton oil and 7,12-dimethylbenz (*a*)anthracene (DMBA) on melanocytes in hamsters. (15 min.)

In golden (agouti) hamsters, pigmented melanocytes of the general body skin are restricted to hair bulbs and occasional networks surrounding pilosebaceous units (F. N. Ghadially and J. F. Barker, J. Path. Bact., 79: 263, 1960). The dermal melanocyte networks are associated with the connective tissue sheaths of hair follicles at approximately the level of the sebaceous glands, and the number of visible networks increases with advancing age. Pigmented melanocytes are lacking in the general body skin of white hamsters (O. Illman and F. N. Ghadially, Brit. J. Cancer, 14: 483, 1960; W. C. Quevedo, Jr., *et al.*, Nature, 189: 936, 1961). In the present study, white and golden hamsters were repeatedly painted with a 1% croton oil-in-acetone solution. Numerous perifollicular networks of pigmented melanocytes appeared in both varieties within 15 weeks. However, despite continuing treatment for 26 weeks, melanocyte networks retained their structural integrity, suggesting that the principal action of croton oil is to increase melanin synthesis without significantly affecting melanocyte proliferation (cf. Shubik *et al.*, Cancer Res., 20: 100, 1960). By contrast, 3 applications of DMBA (1% in mineral oil) elicited melanotic tumors in both white and agouti hamsters; by 26 weeks, occasional tumors measured 11 mm in diameter. In agreement with Ghadially and Barker (1960), these tumors were found to arise within perifollicular melanocyte networks, and with increasing size invest numerous hair follicles. Thus, DMBA stimulates increased division and migration of dermal melanocytes as well as increased melanin synthesis.

89

NANCY S. RAFFERTY, Medical School, Johns Hopkins University. Homologous implantation of embryos into adult frogs. (15 min.)

Whole pre-tailbud *Rana pipiens* embryos removed from their membranes were implanted into various sites, including the anterior eye chamber, coelom and subcutaneous areolar tissue, of homologous adults. Depending upon the site, teratomatous growths resulted, as determined by specimens fixed and sectioned at intervals up to one year. Maximum development of the teratomas occurred between the first and third week after implantation in which there appeared neural tubes, notochord, myotomes, pronephros, gut, optic cups and cartilage. After one month, regression of the teratomas had begun, and only masses of pigment cells and cartilage remained. The highest incidence of teratomas and greatest variety of tissues occurred in the anterior eye chamber, the next highest in the coelom and least incidence and variety in the areolar spaces beneath the skin. No malignant growths resulted, in contrast to the experience of G. Andres (Revue Suisse de Zoologie, 57: 1-12, 1950) using *Xenopus*.

However, the embryo implant in the eye was invariably associated with a fibroma-like growth in the host lens for the first three months after implantation. The origin of the cells and their fate are uncertain, that is to say, it is difficult to discern whether they arise from the implant or are induced by the implant. Anterior eye chamber implants of the Lucké renal carcinoma were not associated with a similar growth in the host lens. (Work done under a U.S.P.H.S. postdoctoral fellowship.)

109

C. L. RALPH, University of Pittsburgh. Distribution of cardio-accelerator hormone in *Periplaneta americana*. (15 min.)

Tissue extracts of adult male cockroaches, *Periplaneta americana*, were assayed on the heart of the same species *in vitro* for factors influencing frequency and amplitude of beat. Primary interest centered upon the nervous system, in particular upon the relative content of such factors in the various ganglia, but non-nervous tissues were assayed as well.

A gradient of concentration of cardio-accelerator was found in the nervous system, the greatest amount in the corpora cardiaca-corpora allata (extract produced about 100% increase in frequency), lower amounts in the brain (50%) and subesophageal ganglion (45%) and still less in the thoracics (25% increase by the third one). The abdominals showed a gradient from the first ganglion which had no detectable amount, through increasing amounts in successive ganglia, up to the terminal (or sixth) which produced a 35% increase in frequency. The relative content of hormone

does not reflect merely size difference since ganglia of similar size have different accelerator contents.

The motile visceral structures, gut (particularly midgut), heart, and utricles, and the hemolymph contain cardio-accelerator. Leg and thoracic muscles have little, if any.

The accelerator factor is extremely temperature stable, withstanding 100°C for 15 minutes without loss of activity, but is destroyed by trypsin and chymotrypsin digestion. After one hour of chymotryptic digestion at 40°C a ganglion extract has a reversible inhibitory effect on the assay heart. (Supported by grant G-9813 from N.S.F.)

249

GORDON M. RAMM, University of Maryland. Development of chick embryos under low level gravitational stress.

Chick eggs were incubated continually in an especially constructed centrifuge for the first one to six days of development. The eggs were positioned at radii which resulted in "g" level of 1.1 and 2.2 when spun at 150 R.P.M.

Embryos were examined and classified as normal, within variation limits of controls and abnormal. The abnormal embryos included several types. A percentage of embryos in excess of the expected of the blood-ring type were obtained. Frequently embryos with a normal vascular pattern and active heart exhibited reduced circulating blood or occasionally abnormal membrane formation. Dead embryos were included with the abnormal and may have been the result of any of the above conditions.

Examination of the 1.1 g embryos exhibited little effect during the first 48 hours. Between 72 and 144 hours, abnormal embryos were found from 35% to 50% but in no series did the dead embryos exceed half that percentage. The effects were more extensive in the 2.2 g series with 85% abnormal embryos at 48 hours and 94% by 61 hours. All embryos 72 hours or older were abnormal. The mortality of these series was 8% at 48 hours and followed a straight line regression to 100% by 144 hours.

These results suggest that the early chick embryo is sensitive to continuous low level centripetal stress. Several factors are probably responsible: interference in gastrulation and blood vessel formation, stress on circulating blood and mechanical interference. During the period of treatment the percentage embryos affected is dependent on the duration and quantity of the treatment during the first 72 hours and the time of death is probably related to the type and degree of the abnormality.

87

WILLIE M. REAMS, Jr. and THOMAS C. MAYER, Louisiana State University. The occurrence and migration of pigment cells in the hind limb musculature of PET mice. (15 min.)

Mice of the PET strain consistently possess pigment cells in certain muscles of the hind limb. The origin of these melanocytes has been investigated by the employment of intracoelomic grafting methods to the coelom of White Leghorn chick embryos, and the migratory pathway of the melanoblasts into these muscles examined. Grafts of leg bud tissue including ectoderm from mouse embryos prior to day 12 lack pigment cells, whereas similar grafts from 12½ day old embryos show numerous pigment cells to be present. However, when leg mesoderm is grafted without its covering epidermis, pigment cells are not found in the graft except from donors of 14 days or older. The present study demonstrates that these extraepidermal melanocytes have their origin outside of the hind limb, and migrate from the skin deep into the muscular tissues relatively late in the development of this region. The vast proliferative and migratory capacity of pigment cells within the hind limb musculature in this strain is shown by the large area of the host coelom pigmented from graft tissues of this advanced age. (Supported by N.S.F. Grant G-14153. Junior author a Science Faculty Fellow of the N.S.F.)

60

ERNST S. REESE, University of Hawaii. Inter- and intraspecific dominance relationships of sympatric species of intertidal hermit crabs. (Motion picture, 15 min.)

Intraspecific dominance relationships of crustacea have been reported and reviewed by Bovbjerg (Ecology, 41: 668-672, 1960). However, only one anecdotal account of interspecific dominance has been reported (M. B. Douglis, Anat. Rec., 96: 553-4, 1946).

Shell preferences were established for hermit crabs inhabiting the reef flat at Eniwetok Atoll, the coral cobble intertidal of Kaneohe Bay, Hawaii and the rocky intertidal of the California coast. Pairs of crabs of either the same or different species were made to compete for the preferred species of shell. The crab which successfully defended a shell or evicted another was considered as dominant.

Both inter- and intraspecific dominance relationships were found to exist. Excepting for the unexplained dominance of larger females by smaller females (134 pairs tested), weight was found to be directly correlated with dominance in intraspecific tests. The importance of sex is not conclusive. However, individual differences can override the effects of both size and sex. The importance of initial occupancy is related to the "fit" of the crab in the shell and the aggressiveness of the attacking crab. Continued fighting between the same two crabs resulted in one crab becoming completely dominant, and rarely was the result of the initial encounter reversed in subsequent encounters.

In interspecific dominance all of these factors are secondary to species membership. Again individual differences may overcome the interspecific dominance hierarchy.

86

SHIRLEY E. REISS, Washington University. Growth control of neurons in the superior cervical ganglion of the mouse. (Introduced by Viktor Hamburger) (15 min.)

Superior cervical sympathetic ganglia of mice contain larger neurons in males than in females. They innervate submaxillary salivary glands which also show sexual dimorphism. A proteinic nerve growth factor (NGF) which stimulates the growth

of sensory and sympathetic neurons can be isolated from these glands in amounts ten times greater in males than in females (Levi-Montalcini and Booker '60, Cohen '60).

Castration of male mice at any age causes first reduction in volume of intralobular-duct cells and subsequently decrease in size of large neurons in superior cervical ganglion suggesting that sexual dimorphism of this ganglion is not a direct response to male hormone but reflects differences in size of the peripheral field of innervation.

Accordingly, salivary glands were removed from adult males and females and the superior cervical ganglia studied at regular intervals thereafter. Chromatolysis and cell death followed; by 80 days after operation ganglia were much reduced in size. In males characteristic large cells disappeared first. Thus the superior cervical ganglion, like other nerve centers, is highly sensitive to changes in its peripheral field.

As NGF stimulates excessive growth of sympathetic neurons, the question arose whether its administration could counteract hypoplastic changes in superior cervical ganglia resulting from extirpation of salivary glands. A number of desalivated mice were therefore injected with NGF for two weeks starting at weaning. Results indicate that this treatment does prevent some of the effects of extirpation. Thus the NGF as well as the size and condition of the peripheral field may be considered to control the growth of sympathetic neurons.

134

T. M. RIZKI, Reed College. Experimental analysis of hemocyte morphology. (30 min.)

Two types of spherical hemocytes are found in *Drosophila melanogaster* during larval life: the crystal cells which are characterized by their cytoplasmic crystalline inclusions and the plasmatocytes. At pupation, the hemocyte population consists primarily of cells with filamentous or pseudopod-like extensions and cells which are extremely flattened. These cells are variants of the plasmatocyte class. The morphological alterations of the plasmatocytes can also be obtained during early larval life by manipulation of hereditary factors or by changing physiological conditions through experimental means. Some of the experiments which elucidate the properties of the plasmatocytes and their variants will be discussed. (Supported by grant RG-5285 from the U.S.P.H.S.)

153

JOHN L. ROBERTS, University of Massachusetts. Day to night respiration of carp, *Carassius carassius* (L), following thermal acclimation and photoperiod control. (15 min.)

Previous experiments (E. von Buddenbrock, '60; Roberts, '60) have shown that European carp, *C. carassius*, inversely compensate oxygen uptake to temperature (Type 5 of Precht, '55). The fish also were found to maintain respiration on 17-hour photoperiods at levels significantly above animals on 7-hour photoperiods when acclimated to 20°C (Roberts, '61). At lower acclimation temperatures, 12° and 5°C, differences related to photoperiod were not found. In contrast, respiration of excised gill and muscle from the same fish showed positive thermal acclimation (probably Precht's Type 3), but no clear photoperiod response.

Because of this contrast, continuous, overnight recordings of respiration were made polarographically in an open water-flow system with fish acclimated to 5°, 12° and 20°C and simultaneously adapted to 7- and 17-hour photoperiods. Absolute reductions in respiration levels as well as significant proportional reductions were observed in the inverse thermal-compensation expressed by fishes after dark when compared to daytime. Although the degree of difference in the photoperiodic effect on respiration in darkness showed also an absolute reduction, the proportionality remained unchanged from that of daytime.

The appearance of the photoperiodic effect at only the 20°C acclimation temperature suggests the existence of a critical temperature for *C. carassius* below which significant changes in respiration to photoperiod alterations do not occur. Also, it is suggested that respiratory responses of carp to these environmental factors indicate differences in their seasonal requirements for feeding and reproduction in summer and for anaerobic existence within sediments of pond bottoms during winter. (Supported by grant RG-6377 from the U.S.P.H.S.)

106

KENNETH D. ROEDER, Tufts University. Ultrasonic echoes from flying moths. (Motion picture, 15 min.)

Flying bats approach and investigate a pebble tossed in the air, but usually do not attack it. Acoustic discrimination of such inert objects from the normal prey could be based on criteria such as form of flight path, wingbeat tone, or fluctuations in the strength of the echo due to changes in the wing attitude of the prey. The echoes cast by moths exposed while in fixed flight to a sequence of ultrasonic pulses (60 to 100 kc/sec., 0.6 msec. duration) were registered oscillographically using a Granith microphone as detector. The moth was illuminated stroboscopically at the instant the sound pulse reached it, and its flight attitude photographed on the same frame as the oscillogram. The echo varied by 20 db or more with the angle of the moth's axis to the sound path, and with the aspect presented by its wings to the sound pulse. In the horizontal plane a maximum echo was cast when the moth was oriented approximately 90 degrees to the sound path, and when its wings were close to the top of their stroke. Thus, the wing surface appears to be the major source of echoes. This suggests that a bat receives echoes of its cries that are modulated in amplitude at the wingbeat frequency of its prey, and that when both prey and predator are on a horizontal course insects crossing the bat's path at about 90 degrees are most readily detected. (Supported by grant E-947 from the U.S.P.H.S.)

110

HARRY D. ROUNDS and MARLENE McCLAIN, University of Pittsburgh. Segmental distribution and response of insect chromatophorotropins. (Introduced by Paul A. Wright) (15 min.)

Nervous tissue of the cockroach *Blaberus giganteus* was freeze-dried overnight and extracted with diethyl-ether followed by an alcohol-water series, beginning with 100% ethanol, then 95% ethanol, 85% ethanol . . ., etc. and ending with water. This procedure allowed a sharp separation of the hormones. They appeared in the 100% ethanol, 65% ethanol, 35% ethanol and water fractions. Fractions were assayed *in vitro* on crustacean carapace.

The chromatophorotropins obtained by this procedure depended on the ganglion extracted, light intensity, and background coloration. When the brain of a cockroach kept on a light background under low light intensity was extracted, dispersing hormones were found in the 100% ethanol and water fractions. However, when the brain of an animal kept on a dark background under low light intensity was extracted, concentrating hormones appeared in the 100% ethanol, 35% ethanol and the water fractions.

Each segmental ganglion under specified conditions yielded a characteristic set of hormones. The segments could be divided roughly into two groups on the basis of factors found in the 100% ethanol and the water fractions when the animal has been exposed to specified conditions of illumination. The first group consisted of the brain, the 3 thoracic ganglia and the second abdominal ganglion. These ganglia responded in unison to illumination changes. The second group consisted of the remaining ganglia, *i.e.*, subesophageal, the first abdominal and the third through the sixth abdominal, and did not appear to respond to these changes. (Supported by grant G-9813 from N.S.F.)

250

LAURENS N. RUBEN and JEAN STEVENS, Reed College. Some aspects of homograft-induced accessory limbs in urodeles. I. Systemic dosage effects.

It is generally conceded that implant breakdown plays a role in initiating implant-induced accessory limb formation in urodeles. (Ruben, Amer. Nat., 94). This series of experiments represents an attempt to take advantage of situations revealed by transplantation experimentation. They were designed to study the effect on supernumerary limb induction of (1) increasing the quantity of implant material per host, by utilizing from one to four limbs per host as implant sites, and (2) of transplanting equivalent quantities of implant material at two-week intervals, rather than simultaneously as in (1). All 390 implants were large kidney homografts made subcutaneously into the radio-ulnar and tibio-fibular portions of the limbs of 160 adult *Triturus viridescens* hosts. The experimental period was 70 days postfinal implantation in all cases. All host limbs were prepared for histological examination, and correlations between induced host limb accessory morphogenesis and the condition of the implant were made. Increasing the amount of donor material challenging the individual host fails to increase the percentage of induction, which remained low (from 0 to 1.67%). In addition, it was found that the extent of implant breakdown does not increase with increase in the number of implants per host. Neither did the induction percentages improve by challenging the hosts at two-week intervals, *i.e.*, no summation effect was obtained; nor did more extensive breakdown of the donor implant material result. There is, therefore, no apparent systemic dosage effect when separate limb sites are used, *under these experimental conditions.* (Supported by grant C-2913 from the U.S.P.H.S.)

251

LAURENS N. RUBEN and JEAN STEVENS, Reed College. Some aspects of homograft-induced accessory limbs in urodeles. II. Local dosage effects.

One hundred and forty homograft kidney implants were made into the humeral and radio-ulnar portions of right forelimbs of 70 adult *Triturus viridescens* hosts to study accessory host limb morphogenesis with regard to (1) implant proximity, (2) implant material from the same and different donors and (3) implantation at two-week intervals. The most effective inductive situation (25%) consisted of two implants, from the same donor, made simultaneously. In view of previously reported results, this points to a local rather than a systemic dosage effect. The data further suggest that the same donor was utilized for these particular hosts and that specific donor-host interactions are important. Implants made at two-week intervals, from the same or different donors, induced in only 5% (rather than 25%) of the host limbs tested. Implants made simultaneously from the same donor in the same limb show no greater breakdown than those of previously studied less inductive groups which utilized separate limb sites. However, no implants which were judged to be in good or excellent condition induced in any experimental group. When the two implants were made at two-week intervals the extent of breakdown increased, especially when both came from the same donor. The results suggest a quantitative relationship, since two implants made simultaneously supplement one another, while those made at intervals do not. The factor (s) from the initial implant, then, appear (s) to be reduced by the time the second implant is releasing its factor (s). (Supported by grant C-2913 from the U.S.P.H.S.)

252

GEORGE D. RUGGIERI S.J., MORRIS H. BASLOW, SOPHIE JAKOWSKA and R. F. NIGRELLI, New York Aquarium, New York Zoological Society. Developmental aberrations in sea-urchin eggs induced by sponge extracts.

Antimicrobial and growth-regulating properties of water and ethanol extracts from several species of sun- and oven-dried sponges have been reported (Jakowska and Nigrelli, 1960). Unfertilized and fertilized eggs of *Arbacia punctulata*, in various stages of development, were treated for 5-30 minutes and for 1-6 hours with extracts from the green sponge *(Haliclona viridis)*, heavenly sponge *(Dysidea etheria)*, fire sponge *(Tedania ignis)*, solid sponge *(Cryptotethya crypta)*, bleeding sponge *(Oligoceras hemorrhages)*, sulfur sponge *(Cliona celata)* and bearded sponge *(Microciona prolifera)*.

All extracts were toxic in relatively high concen-

trations (0.5% and 0.05% by dry weight), resulting in cytolysis, interruption of late cleavage, bottom-swimming blastulae surviving for only 48 hours and abnormal plutei; except for the green sponge, development was normal in lower concentrations.

Water extracts from green sponge produced different effects. Two hours after exposure to 0.0001% solution unfertilized eggs developed parthenogenetically, occasionally exhibiting unequal cleavage. Fertilized or unfertilized eggs treated for 5 minutes with a 0.01% solution underwent cytolysis or changes in the jelly coat which caused the eggs to adhere to one another or to the glass surface. Exposure for 5 minutes to 0.001% concentration resulted in a large percentage of cytolyzed eggs and some parthenogenesis. Unfertilized eggs exposed to this concentration for 5 minutes, transferred to fresh sea water and fertilized 10 minutes later, underwent normal cleavage but resulted in many abnormal plutei. Prolonged exposures (4 hours or more) to 0.001% and 0.00005% solutions prevented the development of swimming forms from normally fertilized eggs. (Supported in part by ONR contract Nonr 2266 (00).)

253

ROBERTS RUGH, College of Physicians and Surgeons, Columbia University. Sequelae to low-level X-irradiation of the mammalian embryo.

Mammalian (mouse) embryos which were exposed to as little as 5 r at 0.5 days post-conception (pre-cleavage) showed an increase over the controls of early death and resorption; some delay in cleavage; increase in post-implantation death; and, upon cytological analysis, some embryos exhibited hyperchromatic nuclei, unequal staining of the cytoplasm, swelling of the nucleus and cytoplasm, separation of the blastomeres, sticky and lagging chromosomes. Thus, the very early embryo is probably the most radiosensitive of all mammalian cells.

The common effect of most X-irradiations on the early embryo is protoplasmic and/or cell deficiency. Radiation-killed protoplasm is eliminated. This is later manifested as stunting, microcephaly, microphthalmia, etc. and the central nervous system appears to be the most vulnerable system.

Recent studies on post-natal rats which had been X-rayed in utero give evidence of changes in their electroencephalograms; their electroretinograms; their learning capacities and behaviour. While these effects are not correlated with any morphological changes, they are evidence of permanent functional damage to the central nervous system due to low level X-irradiation of the very early embryo. These are deficient animals due to embryonic irradiation.

254

OLIN RULON, Northwestern University. The extension of fertilizability and life in sand dollar eggs with vitamin B_{12}.

Cobaltous chloride has been found to increase the length of the fertilizable life of eggs taken from the ripe ovaries of *Dendraster excentricus*. When sulfhydril compounds are added to the cobalt in sea water the fertile life may be extended to 25 days. The present work deals with the cobalt compound - vitamin B_{12} (cyanocobalamin) on the fertility of sand dollar eggs.

Five different concentrations of vitamin B_{12} (ranging from 6.25 to 100 mcg per cc) with 0.15% benzyl alcohol (as a preservative) permitted eggs (3-30%) to be fertilized at 96-120 hours after removal from the ovaries while control eggs in sea water could not be fertilized at 48 hours. Benzyl alcohol alone (0.15% sea water solution) had a preserving action on fertilizability but less than when in combination with B_{12} (50% fertilizable at 48 hours, none at 96 hours).

It is suggested that vitamin B_{12} (and perhaps benzyl alcohol) inhibit the decay of fertility by preventing destructive oxidations (*i.e.* sulfhydril to disulfide) at the egg surface.

76

YUICHI SAEKI and KEITH I. BROWN, Ohio Agricultural Experiment Station, Wooster, Ohio. Comparison of fertilizing ability of ejaculated and vasa deferentia spermatozoa in turkeys. (15 min.)

The purpose of this experiment was to compare the fertilizing ability of ejaculated and vasa deferentia spermatozoa.

Fertility tests were conducted three times, using Small White turkeys, during the period September 1960 to March 1961. Both ejaculated and vas deferens semen were obtained separately from several males. The semen samples were diluted with Lake's (1960) solution and inseminated artificially with a single insemination of 0.03 cc per female. The remaining semen was used for observation of sperm morphology. A total of 48 females were provided for each trial, and eggs were set weekly over a 6-week period.

The pooled vasa deferentia semen generally showed higher sperm concentration, survival time and lower percentage of crooked-necked type of abnormality than the ejaculated semen. However, undiluted ejaculated semen showed a significantly ($P < 0.01$) higher fertility than that of the vas deferens. There were no significant differences within diluted semen samples. Spermatozoa obtained from the posterior part of the vas deferens possessed a significantly ($P < 0.05$) higher fertility than those obtained from the anterior part.

These data suggest that there are secretions from the posterior vasa deferentia and at the time of ejaculation that enhance the fertilizing ability of turkey spermatozoa.

255

A. M. SCHECHTMAN and CHARLES C. CHAMBERLAIN, University of California, Los Angeles. Effects of various doses of antigen on suppressed birds.

Within 24 hrs after hatching, chicks were injected with 150 mg of bovine serum albumin (BSA) per chick. This dose was previously found sufficient to induce immunological suppression demonstrable at 45 days of age. In the present birds, for reasons unknown, antibody production was not different enough from that of normal birds (not injected shortly after hatching) to be statistically significant. A trend toward suppression appears evident, and

from our own experience and that of others, it is highly probable that a borderline suppression had been induced.

Experimental and control birds were tested for antibody production by injection of either 10, 40, or 200 mg BSA/Kg body weight at 45 days of age and blood samples were taken at 3, 8 and 16 days after injection. In both experimental and control birds those challenged with 200 mg/Kg produced significantly higher antibody levels than those receiving 10 or 40 mg/Kg.

Mitchison recently presented evidence indicating that chickens remain suppressed so long as the suppressive antigen is maintained in their tissues by repeated injections during embryonic and later life. On the basis of the retention theory one might expect that the present "borderline suppressed" birds would produce lower antibody levels than controls, and that suppressed birds receiving higher challenge doses of antigen would produce less antibody than those receiving lower challenge doses. Hence the present data are interpreted as not supporting the antibody retention theory of immunological suppression. (Supported by grant RG-4466 from U.S.P.H.S. and grant Nonr 233 (40) from ONR.)

256

A. M. SCHECHTMAN and HECTOR TIMOURIAN, University of California, Los Angeles. Enhancing effect of cortisone on antibody production by chickens.

Chicks treated shortly after hatching with 100 mg of human gamma globulin per chick were tested for anti-HGG at 45 days of age by a single injection of 40 mg HGG per Kg body weight. Immunological suppression was clear as shown by comparison with control birds (not injected shortly after hatching). Cortisone at the level of 10 mg/Kg body weight, injected into newly-hatched chicks was followed by markedly higher concentrations of antibody production when the chicks were tested at 45 days of age with the standard dose of HGG. Newly-hatched chicks injected with both cortisone and 100 mg of HGG showed a clear tendency toward immunological suppression but antibody levels were not significantly different, on the basis of the statistical multiple range test, from antibody levels of control birds. Much larger doses of cortisone (200 mg/Kg body weight) caused no greater effect than the smaller dose; it is probable that the larger dose was less effective. Other groups of birds were similarly studied using bovine gamma globulin as antigen. Because of the great variation in antibody production by individual birds, the suppressive effect of the antigen alone and antigen plus cortisone was not statistically significant. However, injection of 10 mg cortisone alone/Kg body weight into newly-hatched chicks was followed by higher antibody production at 45 days of age. These results indicate that at the developmental stage when the chick is susceptible for immunological suppression by antigen, it is also susceptible for immunological enhancement. Moog showed that cortisone stimulated enhanced phosphatase activity within a few days after its injection into advanced chick embryos; whether this enhancing effect would persist for as long as 45 days is not known. (Supported by grant RG-4466 from U.S.P.H.S. and grant Nonr 233 (40) from ONR.)

72

ANTHONY J. SCHMIDT, College of Medicine, University of Illinois. Architectural regularity of the subepidermal reticulum of the adult newt, *Triturus viridescens*. (15 min.)

The histochemistry applied in this study did not reveal any elastic fibers within the subepidermal connective tissue. However, a considerable amount of fibrous collagen and reticulin protein was demonstrated. These latter proteins characterize particularly the adepidermal reticulum (M. Singer and J. S. Andrews, '56), which we have found to be a highly organized structure.

The subepidermal reticulum has a foundation of adepidermal fibers extending in all directions along one plane, not haphazardly, but rather as organized groups of fibers that interlace at intervals forming a nodule-like structure. From this uniplanar foundation fibers extend upwards between the basal cells of the epidermis for a variable but usually short distance. These projections may serve as anchor pegs joining the epidermis with the subepidermal reticulum. Long fibers extend from the adepidermal network deep into the tissue pad. Here, fibrous interconnections could well form a secondary uniplanar network, and in this way enclose the dermis in a fibrous basket. Indeed, the subcutaneous glands, typical for the adult newt, are supported in just such a fibrous basket.

In addition to the protein component, polysaccharides appear to be associated with the fibers of the subepidermal reticulum. Therefore, we should consider the possibility of these latter fibers being composed of, or being associated with, a muco- or glycoprotein complex. (Supported in part from an institutional grant from the American Cancer Society and in part from a grant (RG-6208) from the U.S.P.H.S.)

23

MYRTLE V. SCHNELLER, Indiana University. Genic interrelationships between two particle-bearing stocks of syngen 4, *Paramecium aurelia*. (15 min.)

Killer paramecia that bring about rapid lysis of sensitives carry in their cytoplasm large numbers of Feulgen-positive lambda particles. First found in syngen 8, rapid lysis lambda-bearing killers have now been found in syngen 4, stock 239. This made possible a comparison, by breeding analysis, of the genes of the paramecia required for the maintenance of lambda and kappa, another type of cytoplasmic particle which exists in certain other stocks of syngen 4. The kappa in killers that induce aboral humps on sensitives has long been known to depend for its maintenance on gene K; it disappears rapidly and irreversibly in homozygotes for k. The genes K and k were introduced, by crosses, into the rapid-lysis-producing, lambda-bearing, stock 239. Lambda and rapid lysis killing were maintained both in the presence of the KK and kk genotypes. Hence, the maintenance of lambda is independent of the two alleles at the locus affecting

kappa. The genotype of stock 239, which is adequate for the maintenance of lambda, was tested to ascertain whether it can also maintain kappa. Lambda-free stock 239 animals were used to cross to stock 51 KK kappa-bearing animals. Their F2 showed one-gene segregation of the capacity to maintain kappa. Hence, stock 239 carries either gene k or another gene with comparable effect on kappa. (Supported by grant American Cancer E 81 B to Sonneborn.)

7

J. P. SCOTT, Roscoe B. Jackson Memorial Laboratory. Spectrographic analysis of dog sounds. (15 min.)

Samples of the common sounds produced by dogs were tape-recorded and later analyzed. Barks are usually short sounds with a sharply definite onset and are produced in a highly similar series by any particular individual. It is suggested that these characteristics permit accurate localization of the sound source. In contrast, yelps produced by an isolated puppy are often highly variable; this characteristic may serve to prevent habituation in a listener.

257

CHARLES T. SEHE, State University of Iowa and University of Cincinnati. Radioautographic studies on the ultimobranchial body and thyroid gland in vertebrates: reptiles and birds.

The radioautographic method has been used to study the functional nature of the ultimobranchial body and especially its relation to the thyroid gland. Carrier-free iodine (13-32 μc I^{131}) was administered to selected adult specimens of turtles, lizards, snakes and birds. Radioautographs of ultimobranchial and thyroid tissues were prepared, using the wet-mount method of Evans ('47).

The ultimobranchial bodies were never found in direct contact with thyroid tissues, though, in some individuals, they are frequently located at the caudal level of the thyroid gland. Some similarity to the cellular structure of the parathyroid gland is attained by ultimobranchial tissue in *Chrysemys p. marginata, Pseudemys s. elegans, Coluber constrictor, Euplectes o. franciscana* and *Passer domesticus*. In *Thamnophis sirtalis* and *P. domesticus* the thymus gland is often invaded by cell masses of ultimobranchial origin. The present observations on *Sceloporus u. undulatus* and *Phrynosoma cornutum* are in accord with those reported for *Lacerta agilis* (Eggert, '38): secretory activity of the ultimobranchial gland in these forms is apparently influenced by seasonal changes. In older specimens of reptiles and birds, large epithelial cysts containing granular secretion characterize the gland.

Present experiments confirm that reptilian and avian thyroid glands metabolize and store iodine. Variations in concentration of I^{131} in follicles within the same gland are more apparent in reptiles than birds. Radioautographic preparations of ultimobranchial glands, however, gave no evidence of iodine storage. As reported for fishes and amphibians (Sehe, '60), ultimobranchial bodies of reptiles and birds do not share with the thyroid gland the capacity to metabolize iodine. (Supported by grants from the N.S.F. and the U.S.P.H.S. to the State University of Iowa.)

122

HERBERT F. SEVERSMITH, American University. Cleavage in the brook lamprey, *Lampetra aepyptera*. (15 min.)

Abstract withdrawn.

81

DON H. SHAW and J. R. PATEL, University of Wisconsin. Species differences between the American bison and domestic cattle as determined by heteroimmune hemolysins. (Introduced by Seymour Abrahamson) (15 min.)

Tests were carried out with a reagent obtained by routine immunization of rabbits with bison erythrocytes. The resulting antisera after absorption with cattle cells, was tested against the red cells of cattle, bison, cattle-bison F_1 hybrids and offspring of crosses between F_1 hybrids and cattle (B_1). The data indicated that this reagent detected an erythrocytic antigenic specificity (or more probably, at least two and possibly three specificities) on the red cells of bison which was not present on the erythrocytes of domestic cattle. This includes tests on more than 300 cattle (including a variety of dairy and beef breeds) all of which were negative; and some 400 bison (from two herds), all positive. The erythrocytes of all 13 of the F_1 hybrids tested were reactive. Of the 31 B_1 individuals tested we found 27 were positive while only 4 were negative. This is a highly significant deviation ($p < .01$) from a 1:1 segregation and is consistent with the 7:1 ratio expected for three factor segregation. This study, unlike the earlier work by Owen, Stormont and Irwin (Evolution, 12:102-110,1958), has shown that there are definite differences between

the American bison and domestic cattle demonstrable by means of cross-absorbing heteroimmune antisera prepared in rabbits against bison bloods. Furthermore, the tests with bison-cattle hybrids suggest that there is more than one genetic factor involved.

131

LESTER C. SHELL, University of Dubuque. Comparison of mitotic figure distribution in the central nervous system of normal and colchicine-treated *Rana pipiens* embryos. (15 min.)

Dividing cells were counted in the first three segments of the spinal cord of *Rana pipiens* embryos fixed at Shumway stages 19 and 21, and at eight-hour intervals over a period of forty-eight hours. Embryos, initially at stage 21, kept in three different aqueous solutions of colchicine were also used. All samples contained five animals, which were fixed in Bouin's fluid, sectioned at 10 μ and stained with Ehrlich's acid hematoxylin. Counts of mitotic figures were made at a magnification of 970 diameters using oil immersion objective.

Mitotic activity in stage 21 generally exceeded any shown in stage 19. Ependymal mitoses showed significant differences in being uniformly in excess of the extra-ependymal ones. In both stages the number of mitoses in the dorsal one-half of the cord exceeded those in the ventral one-half in both the ependymal and extra-ependymal sites.

In each of the first three segments, peaks of mitotic activity appeared in both stages. Analysis of data for each time interval did not show appearance of "traveling waves" of mitotic activity.

All concentrations of colchicine gave uniformly higher numbers of dividing cells than for control animals. Extra-ependymal areas showed mitotic figures for all time intervals to be in excess of the ependymal one for colchicine-immersed animals.

The dorsal one-half of the cord showed a higher number of mitotic figures than the ventral one-half. Mitotic figures per segment were uniformly higher in segment one than segment three and the figures in segment two generally more numerous than segment one. (Supported in part by Grant B-398 from the U.S.P.H.S. to Jerry J. Kollros, University of Iowa.)

83

J. K. SHERMAN, University of Arkansas School of Medicine. Functional survival of cells during the formation and dissolution of intracellular ice. (15 min.)

Presence of ice within cells usually is assumed to assure death while the size of ice formations is said to vary directly with the physical injury induced. Evidence will be presented which questions these assumptions.

MCSA and #440 parakeet tumors and C_3H mouse skin were studied. Cell structure before freezing (control), while frozen and after rewarming was correlated with survival on transplantation. Control and rewarmed tissue was fixed in Bouin's fluid and processed routinely. Microscopic sites and appearance of ice artifacts relative to frozen cells were preserved by the modified freeze-drying technique to be described. Tissues were cooled to –75°C and –196°C at rates of from 1°C/min to 50°C/sec and rewarmed at 180°C/min. Tumor transplants were inoculated in and around the pectoralis major muscle of 116 parakeets and followed for 2-4 weeks as to appearance, size and microscopic structure. Using hair color as a marker, ventral to dorsal autografts of 8 mm full thickness circles of skin were made in 138 4-5 week old female mice.

Data showed that (1) nucleus and cytoplasm were sites of ice formation in cooled cells, (2) little or no structural damage occurred in frozen-thawed tumors while skin often was altered noticeably, (3) % survival of tumors frozen slowly (4°C/min) was equal to that of controls, (4) 19% of frozen-thawed skin grafts were successful, (5) contrary to the vitrification theory of survival, % survival varied directly with size of ice artifacts, (6) intracellular ice and structural alteration, therefore, need not be incompatible with survival. (Supported by grant RG-6418 from the U.S.P.H.S.)

118

ALEX SHIVERS, Michigan State University. Studies of the effect of homologous and heterologous anti-jelly sera on fertilization of eggs of *Rana pipiens*. (Introduced by John R. Shaver) (15 min.)

Antisera were prepared by injecting lyophilized egg-jelly material from four species of frogs (*Rana pipiens, R. clamitans, R. sylvatica* and *R. catesbeiana*) into rabbits. By the Ouchterlony technique it was determined that all four species have common components as well as a number of specific components. By adsorbing anti-jelly serum of each species with jelly from the other species, the specific components of each species have been isolated.

Eggs of *Rana pipiens* were treated with antisera against both homologous and heterologous jellies prior to insemination with normal spermatozoa of *R. pipiens*. Treatments with both heterologous and homologous antisera resulted in statistically significant inhibitory effects on the fertilizability of the eggs. The inhibitory effect of the heterologous serum could be removed by treating this serum with either homologous or heterologous jellies. The inhibitory action of the homologous serum could be removed only by treating with the homologous jelly. These results indicate that the specific components of *R. pipiens* anti-jelly serum are inhibitory as are the common components in the heterologous anti-jelly serum. The specific components of the heterologous sera are non-inhibitory.

The effect of antisera prepared against jelly material of other Amphibian genera has also been tested. In cases (Bufo) where a cross-reaction was observed between these sera and *R. pipiens* jelly, inhibition was also observed. No inhibitory action was seen with anti-jelly sera that did not cross-react with the *R. pipiens* jelly (Ambystoma). (U.S.P.H.S. Research Fellow, Division of General Medical Science.)

158

H. S. SIEGEL and P.B. SIEGEL, Virginia Polytechnic Institute. Genetic differences in response of chick testes to gonadotropins. (Introduced by P. C. Holt) (15 min.)

The influence of sire upon the weight response of the testes of day-old chicks to purified FSH, LH and to an avian anterior pituitary homogenate was determined. Significant differences were found between families of paternal half-sibs in response to 10 or 20 μg of FSH. However, although the magnitude of response to 100 or 200 μg of LH was similar to that of FSH, the genetic differences in response were not significant. A significant difference between sire families was also noted when a combined dose of 200 μg LH and 20 μg of FSH was administered. When 5, 10, 20 and 40 μg of FSH were injected into chicks from 5 different sires, it was found that the mean difference between families of paternal half-sibs increased as dosage was increased with a significant difference appearing at the 40 μg level.

Saline homogenates of anterior pituitaries removed from adult males undergoing behavioral treatments were subcutaneously administered to chicks of different sire families. A significant difference in testes weight response was found between sires. These results indicate the importance of genetic control of the sources of stocks involved when chicks are used in the bio-assay of gonadotropins.

59

P. B. SIEGEL and H. L. MARKS, Virginia Polytechnic Institute. Interactions among chickens in different levels of the social hierarchy. (15 min.)

Two flocks, of 170 Leghorn-type pullets each, were assembled at 20 weeks of age. Combs and wattles were removed (dubbed) from 85 females within each flock. Encounters (fights, peck-avoidances, threat-avoidances and avoidances not associated with threats), from 32 to 320 days after assemblage, were recorded for each flock during thirty-two 50-minute periods. Polynomial curves were fitted for these data. In addition roosting and feeding frequencies were noted.

Total encounters within the flocks declined steadily with time and plateaued at about 110 days. Dubbed pullets occupied the lower social positions. Encounters among pullets within the lower positions were significantly greater than those among females in higher positions. Females of higher rank had more encounters with individuals further removed from them in the hierarchy than with their immediate inferiors. Differences between dubbed and undubbed pullets in roosting and feeding frequencies were not significant.

258

J. EMMETT SIMMONS, BERTIE F. ARGYRIS and THOMAS S. ARGYRIS, Syracuse University. Effect of Ehrlich ascites tumor transplants on the mammary duct epithelium of gestating and lactating mice.

Previously we showed that subcutaneous transplants of Ehrlich ascites tumor (EAT) stimulated the growth of mammary duct epithelium of virgin female mice (J. Nat. Can. Inst. 26: 25, 1961). The greatest degree of stimulation was observed two days after tumor inoculation, and only those ducts within 1 mm of the tumor were stimulated. In order to determine to what extent the physiological condition of the mammary gland modified its response to the tumor stimulus, we studied the effect of EAT transplants on the mammary duct epithelium of gestating and lactating mice.

EAT (0.1cc) was inoculated into the mammary gland of mice at 4 and 15 days of gestation and at 14 and 21 days of lactation. Contralateral glands injected with 0.1 cc of sterile saline served as controls. Animals were sacrificed 2 days after inoculation and pretreated with colchicine 6 hours before sacrificing.

Mitotic activity in ducts of tumor bearing mammary glands of 6-day pregnant mice was 6.15% as compared with 1.35% for contralateral controls. The degree of duct stimulation was less, but significant ($P < .01$) in the 17-day pregnant mammary gland. Stimulation consisted of cell hypertrophy and hyperplasia. In contradistinction to the stimulation of the gestating gland by the EAT, there was no significant effect of the tumor on the lactating gland.

The physiological condition of the mammary gland, then, does play a role in its response to tumor growth stimulation. (Supported by grant P-238 from the American Cancer Society.)

259

CARL J. SINDERMANN, U. S. Bureau of Commercial Fisheries Biological Laboratory, Boothbay Harbor, Maine. The effect of larval trematode parasites on snail migrations.

Intertidal gastropods in northern New England participate in relatively extensive seasonal migrations, moving from the upper tide zones in early winter and returning in spring. Study of two species, *Nassarius obsoletus* and *Littorina littorea*, has indicated that parasitization by larval trematodes may retard or inhibit such migrations, with important ecological consequences for host and parasite.

During a study of the avian schistosome *Austrobilharzia variglandis* (Sindermann, Ecol. 41: 785-790, 1960) it was noted that mud snails *(Nassarius)* parasitized by larval schistosomes tended to remain in the high tide zone in autumn after uninfected snails had moved offshore. This provided a source of infection for migratory birds which are the definitive hosts, but also provided a possible limiting mechanism for the parasite, by immobilizing infected snails in the progressively unfavorable environment of the high tide zone in winter.

Recent study of another trematode, *Cryptocotyle lingua*, which parasitizes *Littorina littorea*, has suggested a similar effect of parasitization on host migration. Larval trematode incidence was highest in high-tide-zone snails during late autumn and winter, after most of the summer high-tide-zone population had migrated to deeper water. Conversely, low-tide-zone incidence was highest in spring, after many snails from winter low-tide-zone aggregations had migrated to the upper tide zones. In each case the observed seasonal change in incidence could be explained by immobilization of parasitized snails.

With both species, use of painted marks to distinguish normal and parasitized snails, and monthly examinations of snail movements and trematode incidences over a two-year period, have pointed to definite influence of larval trematode parasites on host migration.

260

CARL J. SINDERMANN, U. S. Bureau of Commercial Fisheries Biological Laboratory, Boothbay Harbor, Maine. Isoagglutination in elasmobranch fishes.

Isoantibodies are common in the blood of higher vertebrates, but have been reported infrequently in teleosts (Cushing and Durall, Amer. Nat. 91: 121-126, 1957) and not at all in elasmobranchs. That this may be due principally to insufficient study was suggested by the finding of strong isoagglutinins in spiny dogfish, *Squalus acanthias,* big skates, *Raja ocellata,* and little skates, *Raja erinacea,* from the Gulf of Maine.

Individual dogfish were found by cross matching and absorptions to possess isoagglutinins for an erythrocyte antigen present in certain other individuals. Fish that carried the antigen did not have the isoagglutinin, and most fish that lacked the antigen had the isoagglutinin. Isoantibodies were not present in sera from nearly full-term or newborn dogfish "pups," but the specific erythrocyte antigen was detectable in the blood of such early-stage dogfish by reactions with isoagglutinins in adult sera.

Sera of big and little skates also contained isoantibodies for specific erythrocyte antigens. Cross matching cells and sera of big skates indicated the existence of two antigens and their corresponding isoagglutinins. Heterologous cross matching of cells and sera from the two skate species suggested that the same or closely related antigens and antibodies were involved in the reactions.

Immunogenetic studies of these and other elasmobranchs, based in part on antigens detected by natural antibodies, offer promise as criteria in determining degree of reproductive isolation of subpopulations, and may provide information about degree of intermixing.

25

JOAN EILEEN SMITH, Indiana University. Purification of kappa particles of *Paramecium aurelia,* stock 51. (Introduced by Sears Crowell) (15 min.)

If a homogenate of 2×10^8 killer paramecia, grown on a 0.25% Cerophyl infusion and *Aerobacter aerogenes,* is centrifuged at 25,000 xG, suspended in 0.01 M potassium phosphate buffer, pH 6.8, added to a slurry of 1.5 g of ECTEOLA in 50 ml of the same buffer, passed through a column, and the eluate centrifuged and cycled through 4 more columns and centrifugations, the pellet so obtained is predominantly kappa. This pellet is suspended in the buffer and added to the top of a freshly packed ECTEOLA column which has been pre-rinsed with phosphate buffer. The average total yield is 10^{10} kappa and 10^8 bacteria. The total number of particles is estimated by Petroff-Hauser counts, and viable bacteria by plating on yeast-beef agar and nutrient agar. That the particles are kappa was indicated by: (1) phase microscope observations, (2) agglutination of the particles with anti-killer serum cross-absorbed with sensitive animals, (3) retention of the specific and characteristic killing action as well as the ability of the particles to infect sensitives as judged by their conversion into hereditary killers, (4) observations that when 2×10^8 sensitives of the same genotype, grown under the same conditions, were passed through the same isolation procedure, only 10^5 bacteria were obtained as estimated by all the methods used for killer preparations. Separation of the two morphological types of kappa, the B particle which contains a refractile R body, and the N particle which lacks the R body, was achieved by sucrose density gradient centrifugation. Infection succeeded only in fractions containing N particles, killing only in fractions containing B particles. (Supported by grant CF-9541-C1 from the U.S.P.H.S.)

261

T. M. SONNEBORN and R. V. DIPPELL, Indiana University. The limit of multiplicity of cortical organelle systems in *Paramecium aurelia,* syngen 4.

Conjugation of a double animal with a single animal, accompanied by cytoplasmic fusion, leads to formation of triple animals reproducing true to type. Likewise, fusion of two singles with a double during conjugation leads to formation of a quadruple. The division processes themselves are progressively slowed as the number of organelle systems increases; division of quadruples takes many hours. No further increase in number of organelle systems seems to be possible for when two oral meridians are closer than about 80° of arc, the kineties between them fail to cleave at fission and much abnormality results. For example, fusion may occur between the two anterior sutures and between the two posterior sutures near the poles. Fusion of posterior sutures at one stage involves partial cytopyge union, resulting in a forked organelle. Fusions increase in successive fissions, proceeding towards the equator. In sum, singles and doubles reproduce well; triples can be maintained; but quadruples can scarcely complete division. Whenever oral meridians closely approach each other, they interfere with the reproductive processes normally occurring at this meridian, leading to misdivisions and fusions of homologous parts. (Supported by American Cancer grant E 81 B to Sonneborn.)

262

CHARLES H. SOUTHWICK and MIRZA AZHER BEG, Ohio University and Aligarh Muslim University. Social behavior of rhesus monkeys in a temple habitat in northern India.

The social behavior of four groups of rhesus monkeys inhabiting a Hindu temple area in Aligarh, India, was studied from October, 1959, to June, 1960. Group compositions varied throughout this period, but in general the group compositions were as follows (adult males, adult females, infants and juveniles respectively): Group 1—9, 12, 4, 15; Group 1B—3, 2, 2, 4; Group 2—7, 10, 8, 9; Group 3—5, 5, 5, 2. During the study period Group 1B developed as a distinct social group from a subgroup of Group 1. The basic cause of fission appeared to be adult male antagonism.

The groups confined their observed movements to an area of 30 acres, with the exception of Group 3, which made frequent excursions of 1 or 2 miles into other sections of the city.

Exclusive territories were not held by any group, but strong group preferences for specific lodge trees and feeding areas did occur. Intergroup fights were common. Group 1 was dominant to all groups. Group 2 was dominant to 1B and 3. Groups 1B and 3 had variable dominance relationships.

The social organization of Group 1 consisted of three primary subgroups. Subgroup A consisted of 2 leader males and 12 to 18 other individuals. Subgroup B consisted of 1 dominant male and 10 to 14 other individuals. Subgroup C consisted of 5 to 7 subordinate males, which ranged more widely than A or C and initiated intergroup fights more frequently.

Sexual behavior was observed in all months except March, with greatest frequency from October to December. The only births observed occurred in April and May. (Supported by grant RG-6262 from the U.S.P.H.S.)

263

CARL CASKEY SPEIDEL, University of Virginia. Induction by ultraviolet irradiation of variations in the micronucleus of *Tetrahymena corlissi*.

Tetrahymena corlissi is provided with a large single micronucleus, a genetic structure rich in DNA. Cultures of this ciliate were exposed to repeated severe 2537 Å ultraviolet irradiation. Ultraviolet quanta of this wave length are highly absorbed by nuclei acids. Each treatment was severe enough to kill most of the tetrahymenae. Survivors were allowed usually to recover and multiply for about 12 days before the next treatment. From one strain (no. 1) the surviving progeny after 5 ultraviolet treatments consisted largely of individuals containing no micronucleus. Several clone cultures of such amicronucleate tetrahymenae were established and observed for many months. From another strain (no. 33) additional clones of amicronucleate tetrahymenae were obtained after 6 ultraviolet treatments over a 75-day period. Seven treatments of a third strain (no. 6), however, failed to induce any appreciable number of amicronucleate individuals. Amicronucleate tetrahymenae from nos. 1 and 33 remained amicronucleate. They exhibited no conspicuous differences from their normal micronucleated progenitors in structure, swimming and feeding behavior and in reproduction by binary fission. They seemed less viable, however, and were somewhat less aggressive in invading tissues of living injured tadpoles as facultative parasites. From ultraviolet-rayed strain no. 1 also was obtained a clone of bimicronucleate individuals. After remaining predominantly bimicronucleate (75-95%) for about 21 months, they were replaced almost entirely (98%) by the normal ancestral unimicronucleate type. Another clone from strain no. 1 consisted of tetrahymenae with small-sized micronuclei. These were still present as the predominant type after 33 months. (Supported by grant RG-4326 (C4) from the U.S.P.H.S.)

264

JOSE SQUADRONI, S.J. and ALEXANDER WOLSKY, Fordham University. Direct demonstration of sexual differences in skin homografts of the newt *Triturus viridescens*.

Differences between male and female skin homografts in *Triturus viridescens* were previously reported (Pizzarello and Wolsky, Anat, Rec., 131: 589, 1958; Ann. N. Y. Acad. Sci., 87: 45, 1960). A direct and conclusive proof that these are due to sex-linked (or sex-limited) genetic differences in the grafts is now offered. Experiments were performed in which one male and one female skin portion were grafted simultaneously on the same host in symmetrical position in close vicinity. The male grafts were always rejected, the female ones always retained in a great number of experiments, both by male and female hosts. That is, the male host rejects the skin of its own sex but retains that of the opposite sex, while female hosts accept the skin of their own sex and reject that of the opposite one. Color differences in the skin of different body regions were utilized to obtain greater precision in location and observation of the subsequent fate of the grafts without sacrificing the hosts (lighter colored belly skin was grafted on olive colored lateral body wall, interrupting the red and black lateral line: in successful grafting the line remains interrupted, in rejection the continuity of the line is reestablished as the graft is gradually resorbed). This demonstration eliminates any possible differences in the environment that could account for the differential response and greatly reduces the probability of the suggestion that genetic diversity of hosts and grafts, due to random breeding of the experimental animals might be responsible for it.

265

BENJAMIN D. STINSON, Princeton University. Differential effects of normal autografts of whole versus lateral half forearm regenerates on X-irradiated upper arms of adult newts.

Previous investigations (Stinson, Anat. Rec., 136: 285, '60a; Anat. Rec., 137: 395, '60b) have suggested the existence of qualitative differences in morphogenetic capacity of blastemata formed on irradiated limbs implanted with normal autografts of *complete* versus *lateral half forearm regions* taken from advanced limb regenerates, but conclusive verification of the apparent differences was needed.

Both forelimbs of 120 adults *(Triturus viridescens)* were amputated through the distal humerus followed by local irradiation (4,000 r) of left limbs 56 days later. Surgical procedures were employed to test the response of irradiated upper arms to autoplastic implants of (1) irradiated complete forearm regenerates, (2) non-irradiated complete forearm regenerates and (3) non-irradiated radial half forearm regenerates. Appropriate control and irradiated upper arms were reamputated through the distal graft sector three weeks later and observed for 70 days.

Non-irradiated right limbs produced normal 4-digit regenerates despite various surgical insults. Irradiated autografts failed to elicit regenerates from irradiated stumps (40 cases), but *normal* autografts elicited regenerates in 75% of cases (60/80 irradiated stumps). Complete forearm grafts resulted in complete 3- and 4-digit hand regenerates in 44% of successful cases, whereas radial half forearm grafts elicited half hand regenerates with a maximum of two digits in 40% of successful cases.

Both the asymmetry and the skeletal pattern of the normal autograft were reproduced in the regenerate.

These findings demonstrate a correlated variation in graft composition and maximal morphogenetic capacity of the resulting blastema which supports the probable derivation of the regenerate directly from the graft. (Supported by Postdoctoral Research Fellowship (CF-5504-C4), U.S.P.H.S.)

140

FRANK STOCKDALE, JOAN ABBOTT and HOWARD HOLTZER, Medical School, University of Pennsylvania. Behavior of cartilage cells *in vitro*. (15 min.)

What will post-mitotic, differentiated 10-day chondrocytes do when removed from their normal environment and cultured under different conditions for varying lengths of time? Chondrocytes were liberated from matrix by trypsin and reared in a monolayer or in pellets. DNA synthesis was followed radioautographically by means of tritium-labelled thymidine.

Ninety-five percent of the freshly liberated chondrocytes grown in monolayers synthesize DNA in the first 30 hours; they continue to divide *rapidly* thereafter. In addition to demonstrating activation of DNA synthesis in differentiated cells, these experiments indicate that virtually all cells of the original population divide and that their subsequent behavior cannot be attributed to selection of a small group of cells. After approximately a 4-fold increase in cell number over 7 days the progeny of chondrocytes fail to form recognizable cartilage when organ cultured; *i.e.*, they have dedifferentiated.

Approximately 30% of freshly liberated chondrocytes grown in pellets synthesize DNA in the first 30 hours; they continue to divide *slowly* thereafter. A 4-fold increase in number of cells grown in pellets over 21 days does not result in the dedifferentiation of all the cells. Slowly dividing cells in a chondrogenic environment are not as readily dedifferentiated as rapidly dividing cells in a non-chondrogenic environment.

Dedifferentiated cells may differentiate into fat cells and differentiate back into cartilage cells when mixed with freshly liberated cartilage cells. A millipore filter between dedifferentiated and freshly liberated cartilage cells blocks the interaction. Dedifferentiated cartilage cells are not readily incorporated into myotubes. (Supported by a grant B-493 from the U.S.P.H.S. and grant G-14123 from the N.S.F. H. H. is a Scholar in Cancer Research of the American Cancer Society.)

42

LORNA P. STRAUS and W. L. DOYLE, University of Chicago. Fine structure of the so-called chloride cells of the gills of the guppy. (15 min.)

Effects of experimental modifications in the environment on the fine-structure of the so-called chloride cells in the gill are examined in relation to the assumed function of these cells in chloride regulation. *Lebistes sp.* were reared in the laboratory and subjected to variations in temperature, oxygenation and salinity. Attention was directed to the morphology of the endoplasmic reticulum which is present as a branching, smooth-surfaced tubular system. It is shown that the tubules are continuous with the lateral cytomembranes and form distinctive close-packed arrays, net-like patterns and also come into close association with, while remaining distinct from, the numerous prominent mitochondria. Cells of the fresh-water as well as saline-adapted specimens are characterized by the presence of a relatively large apical pit. In the cytoplasm near the surface of this pit small vacuoles are abundant and some of them are shown fusing with the surface. (Supported by grant C-3973 from the U.S.P.H.S.)

266

FREDERICK N. SUDAK and WALTER B. ESSMAN, Albert Einstein College of Medicine. Memory and problem solving after reanimation from deep hypothermia.

The body temperatures of mice were reduced to between 0°C and 5°C (using hypercapnic anoxia) after learning a simple sensori-motor response in a water maze. Reanimation was effected with warming and positive pressure with 95% O_2, 5% CO_2. Control animals were exposed to cold air and then placed in warmed cages. Following achievement of colonic temperature within normal limits all animals were tested for retention of the maze response. Retention was equivalent for both groups with no impairment obvious in the experimental animals. Exposure to cold air (2°C) for 30 min. at successive intervals showed that experimental animals were unable to regulate body temperature over a four-week period, but by the fifth week had regained thermoregulatory ability. Memory of the maze response over this period was unaffected, even with reduced body temperature (T_A range = 4.4°C). Problem-solving ability tested 5 weeks after reanimation by habit-reversal in the maze showed that the control animals were superior to the experimentals ($F = 13.78$; $p < .001$) in their rate of acquisition of the new habit. The results suggest that while initial deep hypothermia and subsequent moderate hypothermia have no effect upon memory, a proactive deficit in problem-solving ability is produced. (Supported in part by Grants H-3536, H-3836 and 2M-6418 from the U.S.P.H.S.)

191

RICHARD P. SUDDICK and THEODORE J. URBAN, Creighton University. A change in acidophilic cells of the pituitary gland of the rat after salivariadenectomy. (15 min.)

There have been many reports that the salivary glands are associated with the endocrine system, or that these glands have an endocrine function themselves. It has been suggested that salivary gland extirpation may cause general inhibition of the anterior pituitary. This study is concerned with changes that occur in the pituitary gland of rats after salivariadenectomy and during administration of salivary gland homogenates. These changes were determined on a quantitative basis by cell counts and qualitatively by histological examination using the modified Martin's technique.

Salivariadenectomy was followed by a significant

decrease in percentage of acidophilic cells and a significant increase in percentage of chromophobic cells from percentages of these cell types in pituitaries of control animals. No significant differences were noted in percentages of basophilic cells.

Histologically, a decrease in cell size, a decrease in cytoplasmic staining intensity, and darker-staining more angular nuclei were noted in the acidophilic cells of the salivariadenectomized group. Nuclei of the basophilic cells were smaller and stained more intensely than those of the controls. The cells were smaller and more angular in shape. Vascularity of the pituitaries of salivariadenectomized rats appeared to be less than that of the controls.

Salivariadenectomized animals that received injections of salivary gland homogenates following the operation and during the course of the experiment did not show these quantitative or histologic changes. (Supported by grant FD-440 from the U.S.P.H.S.)

6

WILLIAM N. TAVOLGA, American Museum of Natural History. Mechanisms of sound production in the marine catfishes, *Galeichthys* and *Bagre*. (15 min.)

A morphological study of the skeleton and musculature in these forms (Family Ariidae) reveals an "elastic spring" mechanism basically as described by Johannes Müller in 1842 for other siluroids. The elastic spring is a lamina of bone derived from the anterior ramus of the parapophysis of the fourth vertebra, herein named the "Müllerian ramus." The protractor muscle that activates this sonic apparatus is innervated by the occipital nerve, and appropriate pulsed stimulation of the nerve can induce sound production which approximates that of a normal fish. Analysis of the sounds produced under natural and artificial conditions and in anechoically controlled situations shows that the fundamental pitch emitted is equivalent to the frequency of contraction of the protractor muscle. Experiments also indicate that the swim bladder functions as an impedance matching device rather than an amplifier or resonator for sounds produced by the "elastic spring." (Supported by O.N.R. Contract Nonr 552 (06) NR 301-322.)

112

ROBERT S. TEAGUE and JANE R. PATTON, University of Alabama Medical Center. An *in vivo* assay of melanophore hormone. (15 min.)

A method for the assay of melanophore hormone (intermedin) using hypophysectomized frogs has been designed and evaluated. Although several species of frogs have been used successfully, best results have been obtained with *Hyla cinerea*. The reflectance of a small area of the dorsal surface of the anesthetized frog is measured on the Beckman Model B spectrophotometer equipped with reflectance attachment and photomultiplier. Samples of hormone contained in 0.1 cc are injected into the ventral lymph sac *via* the hind limb and the reflectance change of the dorsal surface of the frog is recorded as a function of time. A smooth curve is drawn through the experimental points and the peak response read from the curve.

In order to perform a complete four-point assay, four hypophysectomized frogs are each given 2 injections on each of 2 successive days. The order of injections follows a Latin Square design. Using this procedure the relative potencies have been obtained of USP Posterior Pituitary Reference Standard, USP Corticotropin, Li's sheep α-corticotropin, bovine β-MSH, and Lerner's α-MSH. (Supported by N.I.H. grant C-3101.)

13

C. RICHARD TERMAN, Penrose Research Laboratory, Zoological Society of Philadelphia. Behavioral factors associated with the cessation of growth of laboratory populations of Prairie Deermice. (15 min.)

Six populations composed of 4 bisexual pairs of deermice were released into circular pens enclosing 10 or 20 square feet of area. Each population was maintained until at asymptote (no young born for 15 weeks) and food, water and nest boxes were supplied in excess of utilization.

In general, the proportion of each population found in a single aggregate (contactual behavior) during daily examinations was inversely related to the rate of population growth.

Food hoarding began in each population prior to cessation of reproduction. The hoard was not defended and other mice of the population had access to the food. Competition among the mice for the food was not over which animal ate the food, but rather over the place where the food was eaten or hoarded. Any member of a population was permitted to eat the food undisturbed, provided it did so at the location of the hoard.

The data suggest that increased contactual behavior reflects factors operating to cause cessation of population growth. Hoarding behavior, beginning as it does prior to population asymptote, may indicate increasing intraspecific competition or social pressure correlated with changing social or physiological relationships, or both. (Supported by fellowship MF-7891 and by grant H-4759 from the U.S.P.H.S.)

102

E. TOBACH, T. C. SCHNEIRLA, R. LAUPHEIMER and K. GROSSMAN, American Museum of Natural History. The use of a computer in a study of the behavior of rats during the litter period. (15 min.)

The Aronson keyboard consists of a bank of microswitches, each switch in circuit with a pen writing on an Esterline-Angus reaction recorder. Each switch and pen may be assigned to a behavioral item or pattern and a written record of the frequency, duration and sequence of behavioral acts is made by an observer. This apparatus was modified by feeding the electrical impulses into a data collection and storage device (Laupheimer Data Converter) which transmits 36-bit words at the rate of one. two or three words per second to a tape-punching machine. The punched tape, representing a record of the frequency, duration and sequence of behavioral acts can then be reduced and analyzed by

an IBM 1620 or 700-series computer. Data collected and analyzed in this manner will be presented, showing the relationship between female and pups in stable and unstable nest environments.

132

MARCELLUS E. TONEY, Jr., Virginia Union University. The morphology and origin of blood cells of several Crustacea. (30 min.)

Histological characteristics of blood cells of adult male and female crayfish *Cambarus bartoni*, blue crab *Callinectes sapidus* and lobster *Homarus americanus* were studied in the living state by the use of the phase microscope, and in fixed and stained preparations. In addition fixed and stained blood cells were examined in heart and lymphatic tissues. All blood cell types observed in the living state were identified in the fixed and stained preparations.

Cell types were observed in lymphatic tissue and in some blood spaces that were not detected in the unfixed and unstained preparations or in the stained blood cells of the heart. These were intermediate types showing variations in size, form, and staining characteristics, and more or less lymphoid morphological features. It is possible that lymphoid cell types in lymphatic tissue represent the stem cell for differentiation of all other cell types observed in these Crustacea.

119

ALBERT TYLER, ALBERTA SEATON and JACQUES SIGNORET, California Institute of Technology. Further analysis of anti-developmental action of antisera against fertilizins and against other egg-extracts of sea urchins. (15 min.)

It has been shown previously that development can be blocked in fertilized eggs of sea urchins by treatment with antisera prepared against fertilizin or against saline extracts of jelly-less eggs (Tyler, A., Exper. Cell Res., Suppl. 7: 183-199, 1959). The antisera against fertilizin were generally found to be the more highly effective for such purpose; those against sperm and against other tissues of adult sea urchins were ineffective. Strong antisera can block cleavage within one-quarter of a division cycle.

Current studies with fluorescein-labelled antibodies against the fertilizins of the sea urchins *Lytechinus pictus* and *Strongylocentrotus purpuratus* show the presence of fertilizin-like antigens at the surface of the fertilized as well as unfertilized egg. The fertilization membrane does not show marked fluorescence. Regression of the surface fluorescence indicates decrease in these antigens during development.

Antisera prepared against sea urchin egg extracts exhibit many (8 or more) precipitation bands when interacting with the homologous antigen. Removal or decrease in amount of one or two of these bands can be effected by absorption of the antisera with homologous sperm, without appreciable decrease in the development-blocking action. Absorption with fertilizin-coated sperm removes, or diminishes, another band and also removes, or diminishes, the anti-developmental action of the antisera.

The evidence indicates that anti-developmental action, whether by antisera against egg extract or by antisera against fertilizin, is due to antibodies against fertilizin-determinants. It supports further the view that fertilizin-like material is present in the plasma membrane (not the fertilization membrane or vitelline membrane) of the fertilized egg.

267

WALTER S. TYLER, RICHARD F. McLAUGHLIN, Jr. and ROBERT O. CANADA, University of California, Davis. Branches of the bronchial artery to the interalveolar septa of the horse lung.

Examination of serial 2-mm. thick sections of latex-injected lungs revealed branches of the bronchial artery which directly supplied interalveolar septa. The latex was traced in the serial sections from the site injection at the aortic orifice of the bronchial artery to the branches which supplied the interalveolar septa. The relationship of the bronchial artery to the other vessels and lung structures was observed. Histological preparations of selected subgross slices were made to determine the precise localization of the latex. The bronchial artery was observed to supply the bronchial lymph nodes, bronchi, bronchioles, pleura, interlobular septa and vasa vasorum of both the pulmonary artery and pulmonary vein. Branches of the bronchial artery to the interalveolar septa arose from bronchial arteries which supplied the terminal bronchioles, interlobular septa, pleura and vasa vasorum of the pulmonary artery. Similar branches of the bronchial artery to the interalveolar septa of human lungs were observed by Le Fort in 1858 and by Cudkowicz and Armstrong in 1951. Van Hayek in 1945 reported that the ramus alveolaris of the "Sperr" arteries could contain arterial blood from the bronchial artery. The significance of these findings will be discussed. (Supported in part by contract UC-NONR 222 (51).

268

WALTER S. TYLER, JACK PANGBORN and LOGAN M. JULIAN, University of California, Davis. Fine structure of the air capillaries of the avian lung.

Numerous investigators have studied the system of bronchi of the avian lung and have theorized on the circulation of air within the bronchi and air sacs. The structure of the air capillaries and the mechanisms for ventilation of these structures have received little attention. Portions of lungs from 8- and 17-week old Single Comb White Leghorn chickens reared under strict isolation and free of all known diseases were prepared for electron microscopy by fixation for 2 or 4 hours in Palades solution of osmium tetroxide followed by embedding in methacrylate. Sections were cut with a Porter-Blum microtome using glass knives. Sections approximately 1μ thick were stained by PAS, H&E and toluidine blue and were studied by light microscopy. Thin sections were examined with an RCA EMU-3E electron microscope. The nucleated erythrocytes were readily identified by their characteristic morphology and osmophilic nature. The vascular endothelial cells had microvilli and numer-

ous small vacuoles which may indicate pinocytotic activity. The epithelium lining the air capillaries appeared complete and was thinnest in areas adjacent to blood capillaries. A basement membrane was observed. Thus epithelium, basement membrane and vascular endothelium separate the air from the blood in the air capillaries of the avian lung. No differences were observed in the ages studied. Macrophages were identified by phagocytized colloidal thorium dioxide using methods similar to those of Low and Sampaio (Anat. Rec., 127: 51-63). (The chickens free of known diseases were obtained from the Department of Avian Medicine, University of California at Davis. Supported in part by grant RG-5868 from the U.S.P.H.S. and UC NONR contract 225 (51).)

92

PIERSON J. VAN ALTEN, College of Medicine, University of Illinois. Modification of graft-versus-host reaction in the chick embryo. (15 min.)

Previously it has been observed that splenomegaly and hepatomegaly of the chick embryo occurred following grafts of adult spleen and duodenum to the chorioallantoic membrane (CAM). These changes in the embryo have been attributed to an immune reaction by colonization and proliferation of immunologically competent adult cells, and is referred to as the graft-versus-host reaction. In this study the adult chicken was injected intraperitoneally or subcutaneously with various amounts of 9- and 12-day whole embryonic homogenate. After a suitable time, pieces of duodenum and spleen of the injected adult chicken were placed on the CAM of 9-day embryos and host organs were removed and weighed on the 18th day of incubation. Controls were either spleen and duodenum pieces of uninjected adults or a drop of saline placed on the CAM. When tissues obtained from adult animals injected subcutaneously with embryonic homogenate were grafted, the embryonic host spleen and liver weight did not increase significantly over that of saline controls. On the other hand, when the adult chicken had been injected intraperitoneally with embryonic homogenate its tissues when grafted produced a marked splenomegaly. These findings permit the postulation that following subcutaneous injections of homogenate the adult animal's immunologically competent cells become sensitized to the embryonic antigens and when such cells are placed in the embryonic environment they become damaged or destroyed by exposure to the high concentration of the sensitizing antigen (cf. Tyler, 1960). (Supported by U.S.P.H.S. post-doctoral fellowship CF-6731-C3, National Cancer Institute.)

163

JOHN G. VANDENBERGH, Pennsylvania State University. Birefringence and sudanophilia in starling and red-wing blackbird testes. (10 min.)

The purpose of this study was to examine histochemical changes in the interstitium of the testes during selected portions of the reproductive cycle. Starling *(Sturnus vulgaris)* testes were taken from birds collected in the field and from birds brought into recrudescence in the laboratory by photoperiodic manipulation. Red-wing *(Agelaius phoeniceus)* testes were removed from birds collected in the field from April 2 to August 3.

Sections of testes from both species were stained with Sudan B and examined for lipids. Changes in sudanophilia and birefringence correlated well with seasonal behavioral changes in the red-wing. Birefringent and sudanophilic material was plentiful (arbitrary birefringence index of 4.5) in the interstitium early in the season prior to territorial defense, moderate ($BI = 2.0$) during the peak of territorial defense and absent after the breakup of territories. Starling testes, however, were more difficult to interpret due to masking of Leydig cells by the large pigmented cells in the interstitium and the apparent absence of birefringence. An attempt will be made to remove this pigment and to reexamine these tissues.

130

E. PETER VOLPE and SANJOY DASGUPTA, Newcomb College of Tulane University. Development, sex and pigmentary patterns of gynogenetic diploid leopard frogs *(Rana pipiens)*. (15 min.)

Unfertilized eggs of *Rana pipiens* can be activated by sperm of the spadefoot toad, *Scaphiopus holbrooki*. The embryo develops as a gynogenetic haploid, since the foreign sperm does not contribute its hereditary elements (Ting, J. Exp. Zool., 1951). Restoration of the diploid chromosomal complement can be achieved by retention of the second polar body and its subsequent union with the female pronucleus. Suppression of the second maturation division is promoted by subjecting the eggs, shortly after insemination, to heat shock (Briggs, J. Exp. Zool., 1947).

Eggs for induction of gynogenetic diploidy were derived from the burnsi variant (dominant mutant) of *Rana pipiens*. Approximately 8% of the pseudogamously activated, heat-treated eggs developed as gynogenetic diploids. The developmental capacities of these diploid embryos were apparently restricted; only a few developed beyond metamorphosis. Among 11 successfully transformed gynogenetic diploid frogs, 6 were females and 5 were males. The latter were sex-reversed genetic females.

Analyses of the dorsal spotting patterns of the metamorphosed gynogentic diploid frogs revealed that chromatid exchange (crossing-over) occurred between the burnsi locus and the centromere (assuming that the centromere remained undivided during the first meiotic division). Moreover, it had been previously inferred (Volpe, Science, 1961) that modifiers (genes with small effects) alter the magnitude of expression of the burnsi gene. This contention is supported by the results of the present study; the sundry patterns recovered are interpretable on the basis of recombination of modifiers, believed to comprise a linked polygenic system. (Supported by grant G-16317 from the N.S.F.)

71

ROLAND WALKER, Rensselaer Polytechnic Institute. Fine structure of a virus tumor of fish. (15 min.)

Two types of skin warts affect the pike-perch, *Stizostedion vitreum:* besides the well-known lym-

phocystis disease with virus-induced cell hypertrophy, there are also multiple dermal neoplasms associated with a different virus. The two diseases are about equally common in Lake Oneida, and may affect the same fish.

The neoplastic nodules are compact, often spindle-celled, and variable as to vascularization and fibrous support. Electron microscopy shows cytology like that of some virus tumors of warm-blooded animals, with virus particles in abundance. The tumor cells may be closely packed, or may have many pseudopods in interstitial space. Nuclei often have marginated chromatin. The cytoplasm is highly vesicular, and is rich in ribosomes, both in the matrix and on the endoplasmic reticulum.

The virus particles are unlike those of lymphocystis in size, structure and relation to the cytoplasm. The particle is about 1000A in diameter, and has a capsule which is not clearly polyhedral, and a dense core. Complete particles are found outside the cells, between pseudopods and in presumably intracellular spaces of the endoplasmic reticulum; not in the nucleus. They have not been seen in close array. Partly formed particles, like Bittner, leukemia, and some other tumor viruses, show continuity of the capsule with the cell membrane system. Finely filamentous cytoplasm may be found immediately below the peduncles of these particles. (The Micromorphology Laboratory of the N. Y. State Department of Health kindy allowed the use of a Siemens Elmiskop I.)

108

BETTY J. WALL and C. L. RALPH, University of Pittsburgh. A histological study of cockroach neurosecretory cells following dehydration. (Introduced by Ray L. Watterson) (15 min.)

Adult male *Blaberus giganteus* were placed in individual containers under four experimental conditions: A, with dry food and water; B, with water only; C, without either food or water; D, with dry food only. Animals were sacrificed at 3, 5 and 7 days after the start of the experiment, the nerve cord was dissected out, fixed and ganglia were sectioned and stained with either Mallory's triple stain or aldehyde-fuchsin. The sections were examined for effects on neurosecretory cells, the objective being to determine if dehydration results in specific cytological changes in particular cells.

Animals in Group A served as controls and their neurosecretory cells were used as standards for comparing cells from other groups.

In Group B, neurosecretory cells appeared to be completely normal in all ganglia. These animals, starved but given water, served to show that changes found in Groups C and D were not due to nutritional effects.

Group C animals, deprived of both food and water, had by 5 days distinctive, "abnormal" neurosecretory cells, particularly in the supraesophageal ganglion (Nayar, Zeit. Zellforsch., 51: 320) and thoracic ganglia. The neurosecretory cells of the subesophageal and abdominal ganglia appeared to be relatively unaffected. The cells specifically responding to dehydration are the Type I cells (Geldiay, Biol. Bull., 117: 267). Their cytoplasm contains clumps of stainable colloids, the nuclear membrane is indistinct, and the nucleolar boundary is generally crenated.

Group D animals, with food only, presented the same cytological figures as in Group C. (Supported by grant G-9813 from N.S.F.)

269

ARTHUR C. WALTON, Knox College. Amphibia as hosts of nematode parasites. I.

The following records of the presence of parasitic nematodes in amphibian hosts have been added to the catalog of the Parasites of Amphibia since the listing in 1957. ALYTES OBSTETRICANS (Europe): *Oxysomatium brevicaudatum;* ANEIDES FLAVIPUNCTATUS (U. S. A.): *Thelandros (= Oxyuris) dubia;* A. LUGUBRIS (U. S. A.): *Thelandros dubia;* BABINA SUBASPERSA (Japan): *Icosiella sasai;* BOMBINATOR IGNEUS (Europe): *Oswaldocruzia goezei, Oxysomatium schneideri;* BUFO BOREAS (U. S. A.): *Spironoura pretiosa;* BUFO BUFO FORMOSUS (China): *Oswaldocruzia goezei;* B. B. FORMOSUS (Japan): larval *Gnathostoma spinigerum;* B. MARINUS (Bermuda): *Aplectana vellardi, Rhabdias sphaerocephala;* B. MARINUS (Costa Rica): *Ochoterenella digiticauda, Oswaldocruzia subauricularis, Oxysomatium itzocanensis, Rhabdias sphaerocephala;* B. MELANOSTICTUS (China): *Oswaldocruzia goezei;* B. MELANOSTICTUS (East Pakistan): *Aplectana asiatica, Meteterakis govindi, Oswaldocruzia heparia, O. melanosticti;* B. MELANOSTICTUS (Malaya): *Africana singaporensis;* B. VARIABILIS (Spain): *Oxysomatium schneideri;* B. VIRIDIS (Poland): *Oswaldocruzia goezei, Oxysomatium schneideri,* larval Oxyurid of Kolendo, 1959; B. VIRIDIS (Kashmir): *Cosmocerca kashmirensis;* B. VULGARIS (Poland): *Cosmocerca ornata, Oswaldocruzia goezei, Oxysomatium schneideri, Rhabdias bufonis;* DESMOGNATHUS F. FUSCUS (U. S. A.): *Oxysomatium americana;* DICAMPTODON ENSATUS (U. S. A.): *Thelandros dubia, Rhabdias* sp? of Lehmann, 1960; ELEUTHERODACTYLUS JOHNSTONEI (Bermuda): *Aplectana, Cosmocerca* and *Thelandros* spp? of Williams, 1959; E. LUTEOLUS (Bermuda): *Pharyngodon armatus;* EURYCEA B. BISLINEATA (U. S. A): *Oxysomatium americana;* GYRINOPHILUS P. PORPHYRITICUS (U. S. A.): *Oxysomatium longicaudata;* HYLA ARBOREA (Poland): *Oswaldocruzia goezei, Oxysomatium schneideri;* H. A. JAPONICA (Japan): larval *Gnathostoma spinigerum;* H. NASICA (Brazil); *Pteroxyascaris caudacutus;* HYLOIDES GÜNTHERI (Brazil): *Cosmocerca brasiliense;* HYNOBIUS NAEVIUS (Japan): larval *Gnathostoma spinigerum;* LEPTODACTYLUS OCELLATUS (Brazil): *Oxyascaris necopinus;* L. PENTADACTYLUS (Brazil): *Pteroxyascaris similis;* and L. SIBILATRIX (Brazil): *Oxyascaris necopinus.*

270

ARTHUR C. WALTON, Knox College. Amphibia as hosts of nematode parasites. II.

LISSOTRITON PUNCTATUS (Poland): *Oswaldocruzia goezei;* PALUDICOLA DIPLOLISTRIS (Brazil): *Oxyascaris necopinus;* PELOBATES FUSCUS (Poland): *Oswaldocruzia goezei;* PLETHEDON GLUTINOSUS (U. S. A.): *Oxysomatium lon-*

gicaudata; RANA CANCRIVORA (East Pakistan): *Amplicaecum ranae, Camallanus nodulosus;* R. CATESBIANA (Japan, introduced): larval *Gnathostoma spinigerum;* R. CYANOPHLYCTIS (India): *Valariocephalus trilokiae;* R. ESC. ESCULENTA (Poland): *Oswaldocruzia goezei, Oxysomatium schneideri;* R. ESC. RIDIBUNDA (Europe): larval *Contracaecum longicaudatum, Cosmocercella banyulensis,* larval *Gnathostoma hispidum, Oswaldocruzia goezei;* R. ESC. RIDIBUNDA (Turkestan): larval *Gnathostoma hispidum;* R. LIMNOCHARIS (Japan): larval *Gnathostoma spinigerum;* R. MASCARENIENSIS (Africa): *Aplectana courdurieri;* R. NIGROMACULATA (Japan): larval *Gnathostoma spinigerum;* R. PALMIPES (Brazil): *Subulascaris falcaustriformis;* R. PIPIENS (U. S. A.): *Cosmocercoides dukae;* R. RUGOSA (Japan): larval *Gnathostoma spinigerum, Oswaldocruzia goezei;* R. SYLVATICA (U. S. A.): *Oxysomatium americana;* R. TEMPORARIA (Poland): *Oswaldocruzia goezei, Oxysomatium schneideri;* R. TIGRINA (East Pakistan): *Aplectana agubernaculum, A. asiatica, Amplicaecum cacopi, A. ranae, Camallanus thapari, Oswaldocruzia melanosticti, Rhabdias ranae;* RHACOPHORUS spp? of Chabaud & Brygoo, 1957 (Madagascar): *Falcaustra golvani, Harentinema ambocaeca;* SALAMANDRA ATRA (Poland): *Oswaldocruzia goezei;* S. MACULOSA (Poland): *Oswaldocruzia goezei;* S. SALAMANDRA (Poland): *Oxysomatium filiformis;* TRITURUS CRISTATUS (Poland): *Oxysomatium schneideri;* T. CRISTATUS (Hungary): *Capillaria hepatophila;* T. PYRRHOGASTER (Japan): larval *Gnathostoma spinigerum;* and T. VULGARIS (Poland): *Oswaldocruzia goezei.*

271

ALLYN J. WATERMAN, Williams College. Thyroid gland of *Scyliorhinus (Scyllium) stellare.*

In studying embryonic and adult thyroid glands of oviparous *Scyliorhinus* (July and August) flaps were cut in egg cases containing embryos 1.5-13.7 cm in length, weighing 0.4-12.2 g, and tracer I^{131} injected into yolk sac or intraperitoneally. Largest embryos were near hatching. Cases were attached to calcareous red algae at depths around 100 feet, temperature difference around 10°C. Albumen disappears and egg cases change physically with time. Embryonic stages indicate that spawning occurs throughout most of the year. Material was preserved for study. Mortality was highest in embryos removed from the cases and kept in running seawater; temperature and salinity are factors to examine. After 6 hours discrete thyroids of 7-11.5 cm embryos showed radioactivity of 0.18-0.26% of injected dose (scintillation counting); 0.19-0.31, 18 hours; 0.76-0.86, 24 hours; 0.34-2.3, 48 hours. Embryos of 4.0-6.5 cm showed 0.11-0.18% at 18 hours; 0.13-0.90, 24 hours; 0.18-0.34 at 48 hours. Embryos of 5.7 cm were first to show radioactivity (0.2%) at 6-8 hours and 3-cm embryos (0.19%) after 18 hours, attributable partly to state of yolk sac circulation. 3-cm embryos gave positive radioautographs earlier, not 2-cm stages. 12.2-13.7 cm embryos (intraperitoneal) contained 0.61-2.7% at 18-24 hours; values comparable to adult glands which, in turn, showed 0.12-1.7% at 4 hours; 0.14-0.3, 6 hours; 0.12-0.88, 16 hours; 0.42-0.91, 25 hours; 0.72-2.6 (peak), 45 hours; 0.29-0.58 at 96 hours. (Stazione Zoologica, Naples, Italy. Grant G-7473 from the N.S.F.)

115

JOHN L. WEINERTH, CHARLES R. BATTAGLIA and HULDA MAGALHAES, Bucknell University. The non-essential nature of seminal vesicles in golden hamsters. (15 min.)

Male animals of the white-spotted strain occasionally show unilateral or bilateral absence or reduction in size of seminal vesicles. During the past year ten males were found with reduced or absent seminal vesicles at autopsy, but they had produced a total of 17 litters of 75 young. There was a possibility that the condition of the organs at death was not the same as at the time of breeding.

Operations were performed under anesthesia with sodium seconal on proven sires of strains without urogenital abnormalities. Seminal vesicles were ligated at the base, ablated alone or with coagulating glands, or coagulating glands alone were removed. After two weeks for recovery from surgery the animals were bred with females of the same strain. All operated animals were able to produce litters on the first or second breeding. A total of 96 embryos was observed in 12 litters. The number and distribution of embryos during gestation was felt to be a more accurate indication of fecundity than the number born. Animals in the strain observed show an average litter size at weaning of 9.4. Male hamsters are able to reproduce in the absence of seminal vesicles and/or coagulating glands.

272

ANDREW B. WEISS, SAMUEL A. MATTHEWS and WILLIAM C. GRANT, Jr., Williams College. Some effects of temperature and insulin on carbohydrate metabolism in the frog, *Rana pipiens.*

The effect of temperature and insulin on blood-glucose, hepatic and leg muscle glycogen was studied in *Rana pipiens.* The blood-glucose level of stock animals kept at 16°C was 9.2 mg %. After 3 hours at 21°C this rose to 19.0 mg %; after 4 days at 21°C it showed a further rise to 34.1 mg %. Total hepatic glycogen was 1.09 g/100 g at 16°C and 2.04 g/100 g at 22°C. Leg-muscle glycogen varied from 0.61 g/100g at 16°C to 0.53 g/100 g at 22°C. The results suggest that the animals may convert their glucose and glycogen to some metabolic intermediate at low temperatures.

One unit of insulin (Lilly, Iletin) produced a marked reduction in blood-glucose, as was previously observed by Landsberg in this laboratory. At 22°C the glucose level fell from 20.7 mg % at 1 hour, to 6.1 mg % at 2 hours to negligible traces at 3 hours. Less significant results were obtained with 5 units. With 0.25 units, liver glycogen fell from 2.04 g/100 g to 0.75 g/100 g. One unit had no effect nor was any response observed on leg-muscle glycogen.

84

NORMAN K. WESSELLS, Stanford University. Interactions of dermis and epidermis during embryogenesis. (15 min.)

Embryonic chick shank epidermis (stage 37, separated with 1.5% trypsinpancreatin) fails to differentiate normally and becomes necrotic after three days of culture in a protein-free medium (Wessells, Devel. Biol., in press). Dermis prepared and cultured under the same conditions survives indefinitely and forms an extensive inter-cellular matrix that is collagenase digestible, PAS-positive and resembles that formed in later development *in vivo*.

Recombinations of freshly separated epidermis and dermis display keratohyalin and keratin formation, and slight cornification. This occurs only where the original basement membrane side of the epidermis is in contact with the dermis. Basal cell columnar orientation is maintained in such areas. Elsewhere the epidermis behaves like epidermis cultured alone and dies at three days. In recombinants the dermal inter-cellular matrix forms in regions distant from the epidermis, never between cells close to the epithelium. Recombinations of epidermis with stage 37 thigh muscle display columnar basal cells, limited basal mitoses, but no other normal epidermal differentiations.

When explanted epidermis is separated from dermis by a millipore filter (25μ thick, 0.45μ pore diameter, some cytoplasmic penetration), basal cell orientation is maintained, and keratin-containing subperiderm cells form in some cultures. Only basal orientation occurs when epidermis and muscle are placed in a similar trans-filter situation.

The results demonstrate three phenomena of skin differentiation: (1) orientation of basal cells that occurs when epidermis and mesenchymal tissues are intimately associated; (2) a differentiative effect found in dermis-epidermis combinations but not in muscle-epidermis ones; and (3) an epidermal effect that delays dermal maturation. (Supported by a post-doctoral fellowship from the American Cancer Society, and by N.S.F. Grant G-11709 to Dr. Clifford Grobstein.)

143

RICHARD H. WHITE, Washington University. An analysis of the development of the compound eye in the mosquito, *Aedes aegypti*. (Introduced by Viktor Hamburger) (15 min.)

The compound eye of *Aedes aegypti* differentiates from lateral head epidermis in the last larval instar. Eye formation begins at the posterior edge of the prospective eye region, and spreads as a "wave of development" across the rest of the prospective eye. The wave of development is first expressed as a sharply defined band of mitotic cells, followed by cells in various stages of differentiation.

It has been shown, mainly by transplantation experiments, that the wave of development reflects the passage of a factor, essential to the formation of the eye, across the prospective eye region in a posterior to anterior direction. This factor or condition has been experimentally defined as follows: (1) It is limited to the epidermis; it is not found in the hemolymph. (2) It is absent in front of the band of mitotic cells. (3) It can be made to cross the prospective eye epidermis in any direction, although it normally passes from posterior to anterior. (4) Its effect, or at least its initial effect, is to stimulate mitosis in prospective eye epidermis. (5) Head epidermis other than prospective eye epidermis is not competent to react to the factor, and blocks its passage. (Supported by grant 2G-412 from the U.S.P.H.S.)

22

DOROTHEA WIDMAYER, Indiana University. The detection of two new mutations of hump kappa in killer stock 51, syngen 4, *Paramecium aurelia*. (Introduced by T. W. Torrey) (15 min.)

Two new mutations of hump kappa (51m42 and 51m43) have occurred in killer stock 51, *Paramecium aurelia*. 51m42 was detected among the progeny of an X-irradiated 51 killer; 51m43 later arose in a 51m42 culture.

Animals with 51m42 kappa are weak hump killers. They affect fewer sensitives in a given time interval than do 51 killers. An attempt has been made to determine the basis for this reduced killing ability. 51 killers themselves become weak killers when the kappa number/cell is reduced or when the fraction of B particles (kappa containing R bodies) is small. Reduced particle populations, however, are not responsible for the 51m42 phenotype, since under uniform conditions, both killer stocks have comparable numbers of kappa (ca. 1800) and similar percentages of B particles/cell. Because 51m42 killing particles act as rapidly as those of stock 51 and since one sensitive can be killed by one particle, 51m42 killing particles can be considered as effective as 51 killing particles. Reduced killing apparently results from slower liberation rather than intracellular storage of effective killing particles. Whether this slower rate of release reflects a slower formation of killing particles, presumably from B particles, or the existence of a large fraction of B particles incapable of becoming killing particles, is as yet unknown.

51m43 kappa bearers are resistant to hump killing activity, but are nonkillers. Although these animals contain over 3000 kappa/cell, no B particles are present. This lack of B particles is thought to be responsible for the nonkiller phenotype. (Supported by a U.S.P.H.S. Predoctoral Fellowship from the National Institute of Mental Health and by grant AT (11-1)-235 to T. M. Sonneborn from the A.E.C.)

273

CHARLES G. WILBER and PAUL F. ROBINSON, Kent State University and U. S. Army Chemical Research and Development Laboratories. Role of liver glycogen in swimming performance of white mice.

Small rodents swim for various periods of time depending on water temperature. Factors correlated with swimming time have been reported for guinea pigs. In the present work, mice were forced to swim in water baths at 25° and 35°C respectively. Maximum swimming time was recorded. Samples of liver and muscle were analyzed for glycogen content. Blood lactic acid values for each animal were obtained. The results were compared with those for resting controls held in a room at 23°C. Correlation and regression coefficients were fitted for bath temperature versus liver or muscle glycogen, or blood lactic acid. A statistically significant positive correlation was found between bath tempera-

ture and swim time (t, 17.79; r, 0.99) and temperature and liver glycogen (t, 4.01; r, 0.85). Increased body weight of the mice was associated with greater glycogen percentage in liver (t, 3.83; r, 0.77) and possibly in muscle (t, 2.15; r, 0.56). As swimming time increased liver glycogen decreased significantly (t, 4.86; r, 0.89); no significant change in muscle glycogen or blood lactic acid could be demonstrated. A possibly significant positive relation was found between liver and muscle glycogen (t, 2.37; r, 0.60); no relationship could be demonstrated between either liver or muscle glycogen and lactic acid. Liver glycogen varies with swim time: $y = 175 - 85\ x$, where y is liver glycogen in percent and x is swim time in minutes. Swim time varies with bath temperature: $y = 23 + 0.06\ x$, where y is bath temperature and x is swimming time.

21

FRANCIS H. WILSON and PHILIP N. NIOSI, Lebanon Valley College. Some observations on gastric digestion in the horned owl. (10 min.)

The stomach contents of a young horned owl were found to be strongly acid, no pellets were formed and hair and claws of its prey were found in the feces, indicating the usual digestive processes. In older owls, pellets or uneroded bones were egested from the mouth. The stomach content of the older owls was found to vary from slightly acid to slightly alkaline, which would eliminate the consideration of pepsin as the active digestive enzyme. Feeding experiments and the small lumen of the owl's intestine suggest that in these older owls protein digestion is in the stomach and is brought about by enzymes produced in the pancreas or small intestine and forced up into the stomach. (A faculty student research project.)

47

JOHN W. WINKERT, ALBERT S. GORDON and ELIZABETH L. WINKERT, New York University and New York Medical College. Reticulocyte release patterns following erythropoietic stimulation. (15 min.)

After withdrawal by cardiac puncture of 20% of the blood volume of rats, a biphasic reticulocyte release pattern was observed. The first phase was characterized by a slow discharge of reticulocytes into the circulation and lasted approximately 30 hours. The second phase beginning on the second day after bleeding consisted of an abrupt increase in the rate of reticulocyte release. Since the second phase began after approximately one turnover time had elapsed, it was attributed to the release of cells which were activated in an early phase of development.

Previous observations on reticulocyte responses to human urinary erythropoietic stimulating factor (ESF) were made at relatively long time intervals following injections. To investigate the time course of reticulocyte responses to ESF in greater detail, changes in reticulocyte counts were examined daily at two and at twenty hours after each injection. Nearly all the reticulocytes discharged into the circulation after each injection were released within two hours. This prompt release could not have resulted from growth pressure if ESF acted exclusively upon primitive precursor cells. The results of metabolic studies on ESF action upon bone marrow *in vivo* must be analyzed with regard to possible acute changes in the distribution of marrow cellular elements resulting from reticulocyte discharge. Further investigations are being conducted to determine whether the release of reticulocytes was evoked by ESF itself or by another factor present in the ESF preparation. (J.W.W.—Helen Hay Whitney Foundation Fellow. A.S.G.—Supported by grant H-3357 from the U.S.P.H.S.)

135

GERTRAUDE WITTIG, Entomology Research Division, U. S. Department of Agriculture. Investigations of blood cells in insect pathology. (30 min.)

Insect pathology considers blood cells from two aspects: as a component of the defense mechanism of the insect body, and as cells that can become diseased and show pathologies. Since insect blood cells are able to form tissues and to phagocytose, they can perform repair processes and establish immunity reactions, such as producing pseudoepithelia, encapsulating parasites and foreign bodies, enclosing diseased tissue in nodules; and their phagocytic activity constitutes the cellular immunity of insect blood. Other than the numerical and cytological changes occuring during these processes, insect blood cells may undergo true histopathologies that may be caused either by poisons or infectious diseases. Many insect pathogens appear, sooner or later during the course of infection, in the hemocoele; however, at present only certain groups of microbes that also infect fat cells, or fat cells and other cells of the insect body, have been reported to infect blood cells. Most of them are viruses; a few belong to the rickettsiae.

274

MARIA DE ISSEKUTZ WOLSKY, SUSAN REGAN and ALEXANDER WOLSKY, Fordham University and Manhattanville College, Purchase, N. Y. Differences in the facet number of Bar heterozygotes from various inbred populations of *Drosophila melanogaster*.

Earlier studies on an inbred population from Java revealed morphological and physiological differences in comparison with a standard Oregon-R stock (Mulrennan and Wolsky, Anat. Record 128: 593; McGann and Wolsky, *ibid.* 134: 602; Wolsky and Wolsky, *ibid.* 134: 657). Crosses with Bar showed that in the Java population the wild-type condition is significantly more expressed in the heterozygous females than in similar heterozygotes made with the Oregon-R stock. Counting facet numbers the following data were obtained (20 counts in each case): Java/Bar 448 ± 20.1, Oregon/Bar 292.2 ± 14.4; difference 155.8 ± 25.0. In heterozygote Bar females from another inbred stock (Israel) the facet count was 271.1 ± 13.3. The most common facet number in Bar heterozygotes is $360 \pm$ (Bridges and Brehme). Differences in the facet number of Bar heterozygotes, derived from different populations, have been reported before (Ogaki, Zalokar) and were interpreted in various terms. A cause, hitherto not considered, may be the modifying in-

fluence of adaptive gene complexes ("supergenes"), which sometimes produce heterosis in F_1 "population hybrids" and disappear in their *inter se* offspring due to recombination. This view is supported by (1) the existence of supergene-based F_1 heterosis in the stocks investigated (Mulrennan, unpublished), and (2) a considerable decrease of facet number (to 356.5 ±) in F_2 and F_3 Java/Bar heterozygotes. However, the possibility cannot be excluded that a kind of "isoallely" also exists within the Bar segment of the wild-type X chromosome in different populations. (Supported by grant RG-6713 (C 1) from the U.S.P.H.S.)

161

J. M. YOCHIM and V. J. DE FEO, University of Illinois. Control of the decidual cell response by hormones of the ovary. (15 min.)

Pseudopregnancy was induced in rats (200-250 g) by mechanical stimulation of the uterine cervices during estrus (vaginal cornification). Four days post-estrus, uteri were unilaterally traumatized by scratching the entire length of the antimesometrial surface of the lumen with a burred needle. In rats housed under controlled illumination, optimum results were obtained when the time of traumatization was standardized between 1:30-3:30 PM. Ovariectomy and initiation of daily hormone treatment was also effected at this time. Autopsies were performed 5 days after traumatization. Massive deciduomata (DCR) in intact, pseudopregnant rats weighed 2000 mg., a 1200% increase over non-traumatized horns (NT). Water content of DCR uteri was 84% compared to 80% in NT horns. Ovariectomized rats, injected daily with 2.0 mg. progesterone in combination with 0.0, 0.1, 0.2, 1.0, 2.0 and 10.0 μg. estrone, produced DCR weights of 918, 1000, 1328, 2036, 1404 and 789 mg., respectively. Weights of NT uteri increased in proportion to estrone dosage. Maximum DCR water content of 84% was obtained in rats treated with 1.0 μg. estrone and 2.0 mg. progesterone. Water content of steroid-treated NT uteri ranged between 80-81%. Similar augmentative and inhibitory effects were observed when estradiol was used in combination with progesterone. The effects of relaxin on the responses obtained with estrone and progesterone will be discussed. (Supported by grant GF-8558 (C2) from the U.S.P.H.S.)

19

FRED ZIEGLER, E. HARDIN STRICKLAND and ADAM ANTHONY, Pennsylvania State University. Oxygen electrode measurement of tissue slice and homogenate respiration. (15 min.)

A respirometer for tissue slices and homogenates has been constructed. The device consists of a Clark electrode mounted in a sealed vessel and allows convenient measurement of rapid changes in respiratory rates. This instrument was used to measure respiration of tissues from normal control rats and experimental animals which were debilitated. Respiratory stimulation of both liver homogenates and kidney slices was recorded when succinate was employed as a substrate. Subsequent addition of 100μM 2,4-dinitrophenol stimulated respiration. With the respirometer it is possible to study the respiration of tissue slices and homogenates in the presence of various substrate combinations. The sensitivity of the electrode and its rapid response time allow analyses of respiratory changes which are not detectable with ordinary monometric techniques. (Aided in part by grant RG-5112 (R-2), National Institutes of Health.)

AUTHOR INDEX

Author	Page
Abbott, J.	392
Allen, M. J.	337
Allen, R. F.	337
Allen, S. L.	338
Alvarado, R. H.	338
Anderson, E.	342
Anderson, J. M.	338
Andrew, W.	339
Anthony, A.	355, 358, 400
Argyris, B. F.	389
Argyris, T. S.	389
Auclair, W.	339
Auerbach, R.	339
Bagnara, J. T.	339
Baird, D.	340
Baird, I. L.	356
Bak, I. J.	353
Baker, C. L.	340
Balph, D. F.	340
Banks, E. M.	340
Barbehenn, K. R.	341
Barch, S. H.	341
Barfield, R. J.	349
Barlow, G. W.	341
Barnett, A.	341
Barrett, J. M.	342
Baslow, M. H.	384
Battaglia, C. R.	397
Beams, H. W.	342
Beg, M. A.	391
Benolken, R. M.	342
Bernard, G. R.	343
Beschle, R. G.	347
Birge, W. J.	343, 344
Blauvelt, H.	344, 374
Boatman, J. B.	344
Bond, H. E.	344
Boolootian, R. A.	345
Bovbjerg, R. V.	345
Bovee, E. C.	345
Braddock, J. C.	345
Braddock, Z. I.	345
Breland, O. P.	345
Bremiller, R. A.	346
Breneman, W. R.	346
Briggs, R.	346
Brockway, B. F.	346
Bronson, F. H.	347
Brookes, V. J.	347
Brooks, D. S.	349
Brown, K. I.	385
Cairns, J. M.	381
Calventi, I.	347
Camougis, G.	347
Canada, R. O.	394
Candelas, G. C.	348
Carbaugh, B. T.	348
Chamberlain, C. C.	385
Chavin, W.	348
Chladek, M. I.	376
Clark, P. J.	350
Coleman, J. R.	348
Collias, N. E.	349
Dasgupta, S.	395
Davenport, H. A.	369
Davis, D. E.	347
DeFeo, V. J.	400
DeGennaro, L. D.	349
DeLanney, L. E.	349
DiBerardino, M. A.	349
Dice, L. R.	350
Dilger, W. C.	350
Dippell, R. V.	390
Dornfest, B. S.	350
Dougherty, E. C.	350, 351
Downs, W. G., Jr.	351
Doyle, W. L.	392
Dubbs, C. C.	351
Dunbar, R. S., Jr.	378
Eakin, R. M.	352
Edgar, A. L.	352
Eleftheriou, B. E.	352
Elias, H.	353
Elliott, A. M.	353
Emlen, J. T.	353
Esper, H.	366
Essman, W. B.	392
Evans, D. R.	372
Evans, L. T.	353
Evans, R. M.	354
Farmanfarmaian, A.	354
Farner, D. S.	365
Félix, L.	348
Feng, L. S.	368
Ferral, D. J.	350, 351
Ficken, R. W.	354
Firlit, C. F.	359
Fisher, J. R.	373
Fitzpatrick, D.	351
Fleming, W. R.	354
Foreman, D.	355
Fox, A. M.	355
Fraser, R. C.	355
Frehn, J.	355
French, W. L.	356, 366
Fromm, P. O.	360
Frumkes, P. J.	349
Gardner, P. J.	356
Gilbert, L. I.	360, 368
Gilchrist, R. D.	356

Author	Page
Goldberg, E.	356, 378
Goodge, W. R.	357
Gordon, A. S.	350, 399
Gowen, J. W.	365, 377
Grabowski, C. T.	357
Granath, L. P.	347
Grant, W. C., Jr.	357, 397
Greenberg, F.	370
Griffin, J. L.	358
Grossman, K.	393
Grosso, L. L.	358
Handler, E. S.	350
Harclerode, J. E.	358
Heim, W. G.	359
Hershberger, L. G.	359
Hersher, L.	344
Hickman, C. P., Jr.	354
Hisaoka, K. K.	359, 371
Hoffert, J. R.	360
Hoffman, R. A.	356, 360
Hogue, M. J.	360
Holtzer, H.	392
Hommes, F. A.	367
Horner, B. E.	360
Huddleston, C. J.	360
Humphrey, R. R.	346, 361
Ives, P. T.	361
Jakowska, S.	361, 384
Johnson, C.	366
Johnson, C. E.	378
Johnson, N.	362
Johnson, W. H.	340
Jones, J. C.	362
Julian, L. M.	394
Kallman, K. D.	362
Kalter, H.	363
Kamemoto, F. I.	363
Kaplan, H. M.	363
Katz, F. F.	363
Keister, S. M.	363
Kemp, N. E.	364
Kennedy, J. R., Jr.	353
Kerr, N. S.	364
Kessel, R. G.	364
Kidwell, J. F.	365
Kimball, F.	366
King, J. R.	365
King, R. C.	367
Kirschner, L. B.	338
Kitay, J. I.	365
Kitzmiller, J. B.	356, 366
Kleinholz, L. H.	366
Klopfer, F. D.	366
Klopman, R. B.	366
Koch, E. A.	367
Kosin, I. L.	367
Lahille, F.	372
Lane, C. E.	367
Larimer, J. L.	367
Lash, J.	367
Lasiewski, R. C.	368
Laskey, P. A.	344
Latimer, H. B.	368
Laupheimer, R.	393
Lea, M. S.	368
Lemkey, N.	346, 369
Lhotka, J. F., Jr.	337, 369
Licht, P.	369
Lindeman, V. F.	370
Linton, J. R.	354
Lobue, J.	350
Luce, W. M.	370
McClain, M.	383
McCoy, J. L.	378
Mackin, J. G.	371
McKinnell, R. G.	371
McLaughlin, R. F., Jr.	394
Magalhaes, H.	397
Mahowald, A. P.	371
Malone, T. E.	371
Marks, H. L.	389
Matsumoto, K.	372
Matthews, S. A.	397
May, R. M.	372
Mayer, T. C.	382
Meisner, H. M.	372
Mellon, D., Jr.	372
Merklin, R. J.	373
Mersmann, H. J.	380
Metcalf, J. F.	373
Miyake, A.	373
Monesi, V.	374
Moore, A. U.	344, 374
Moore, R. E.	374
Moriber, L. G.	374, 379
Morrill, J. B.	375
Morrow, J. E.	375
Mueller, J. A.	375
Mulvey, P. F., Jr.	376
Murray, D.	376
Musacchia, X. J.	376
Mutchmor, J. A.	376
Nagabhushanam, R.	377
Nalbandov, A. V.	340
Nash, D. G.	365
Nash, D. J.	377
Nash, T. L.	377
Nigrelli, R. F.	347, 361, 384
Niosi, P. N.	399
Norman, C.	356, 378
Nunnally, D. A.	378
O'Brien, D. J.	363
Ogren, R. E.	378
Olinger, D. M.	342
Olivereau, M.	348
Ortiz, E.	348
Pangborn, J.	394
Parks, M. E.	374, 379
Patel, J. R.	387
Patton, J. R.	393
Payne, R. S.	379

Author	Page
Pence, V. W.	356
Pierro, L. J.	379
Pietsch, P.	379
Pikó, L.	380
Porterfield, I. D.	378
Privitera, C. A.	380
Prutkin, L.	374
Purdie, R. C.	354
Quastler, H.	350
Quevedo, W. C., Jr.	381
Rabinovitz, M. J.	344
Rafferty, N. S.	381
Ralph, C. L.	381, 396
Ramm, G. M.	382
Reams, W. M., Jr.,	382
Reese, E. S.	382
Regan, S.	399
Reiss, S. E.	382
Richards, A. G.	376
Richter, H.	345
Riemann, J. G.	345
Rizki, T. M.	383
Roberts, J. L.	383
Robinson, P. F.	398
Roeder, K. D.	383
Roofe, P. G.	368
Rounds, H. D.	383
Ruben, L. N.	384
Ruggieri, G. D.	384
Rugh, R.	385
Rulon, O.	385
Saeki, Y.	385
Sanborn, R. C.	351
Schechtman, A. M.	385, 386
Scheffel, K.	354
Schein, M. W.	348, 377
Schmidt, A. J.	386
Schneirla, T. C.	393
Schneller, M. V.	386
Scott, J. P.	387
Seaton, A.	394
Segal, E.	376
Sehe, C. T.	387
Seversmith, H. F.	387
Shaw, D. H.	387
Shell, L. C.	388
Sherman, J. K.	388
Shivers, A.	388
Siegel, H. S.	388
Siegel, P. B.	388, 389
Signoret, J.	346, 394
Simmons, J. E.	389
Sindermann, C. J.	389, 390
Slingerland, D. W.	376
Smith, J. A.	381
Smith, J. E.	390
Solberg, B.	350, 351
Sonneborn, T. M.	390
Southwick, C. H.	391
Sparrow, A. H.	361
Speidel, C. C.	391
Squadroni, J.	391
Stadler, J.	365
Stevens, J.	384
Stinson, B. D.	391
Stockdale, F.	392
Straus, L. P.	392
Strickland, E. H.	400
Sudak, F. N.	392
Suddick, R. P.	392
Sweeny, P. R.	358
Tavolga, W. N.	393
Teague, R. S.	393
Terman, C. R.	393
Thompson, C. R.	359
Timourian, H.	386
Tobach, E.	393
Toney, M. E., Jr.	394
Tyler, A.	394
Tyler, W. S.	394
Urban, T. J.	392
Van Alten, P. J.	395
Vandenbergh, J. G.	395
Vásquez, C.	348
Volpe, E. P.	395
Walker, R.	395
Wall, B. J.	396
Walton, A. C.	396
Warren, J. M.	377
Waterman, A. J.	357, 397
Weinerth, J. L.	397
Weisbroth, S. H.	367
Weiss, A. B.	397
Wessells, N. K.	397
Westfall, J. A.	352
White, R. H.	398
Widmayer, D.	398
Wilber, C. G.	398
Wilson, D. E.	345
Wilson, F. H.	399
Wilson, H. A.	378
Winkert, E. L.	399
Winkert, J. W.	399
Winner, J. M.	343
Wittig, G.	399
Wolsky, A.	391, 399
Wolsky, M. I.	399
Yochim, J. M.	400
Yokoyama, H. O.	346, 369
Zamboanga, P. M.	359
Zarrow, M. X.	352
Zeller, F. J.	346
Ziegler, F.	400
Zilliken, F.	367